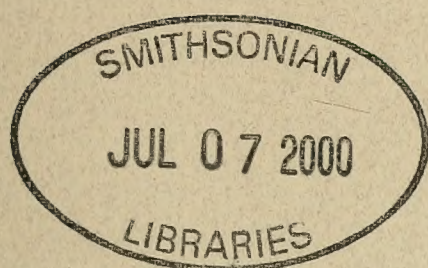


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## **Earthworms (*Oligochaeta: Lumbricidae*) in High-Maintenance and Low-Maintenance Lawns in Lexington, Kentucky**

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and

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### **ABSTRACT**

We sampled earthworms in Lexington, Kentucky, and compared populations in high-maintenance lawns (maintained by professional lawn-care companies) to those in low-maintenance lawns (maintained by individual owners without the use of lawn-care chemicals). We sampled 15 high-maintenance and 15 low-maintenance lawns with similar distributions in terms of geographic location within Lexington, age of house, and length of time the lawn was managed under the current maintenance regimen. Earthworms were collected during the period 6 Apr–10 Apr 1998 in each lawn by using formalin as an extractant. Significantly more earthworms per unit area were collected from high-maintenance as from low-maintenance lawns. Conversely, low-maintenance lawns had significantly greater earthworm dry mass per unit area and dry mass per individual earthworm. These results suggested that high-maintenance lawn care resulted in stunted growth of earthworms. Since earthworms are one of the most important members of the soil fauna, we suggest that organic methods and alternative plant systems be substituted for chemically maintained lawns.

### **INTRODUCTION**

There are more than 25 million acres of lawn in the United States (PLCAA 1999). In 1997, 22% of households employed professional lawn-care companies to maintain their lawns, and the demand for professional lawn care was increasing at an annual rate of ca. 3% (PLCAA 1999). Over 70 million pounds of chemicals are applied annually to lawns with the annual application rate increasing 5–8% per year (Jenkins 1994, p. 186). By the mid to late 1980s, the average lawn owner in the U.S. was using higher concentrations of chemicals

than farmers (Jenkins 1994, p. 186) and more synthetic chemical fertilizers were being applied to these lawns than were being applied to all food crops in India (Jenkins 1994, p. 142).

It is recognized that soil animals in general, and soil invertebrates in particular, are essential to healthy soils (e.g., Brady and Weil 1996). In the eastern U.S., especially in disturbed ecosystems, earthworms (*Oligochaeta: Lumbricidae*) account for a large proportion of the total soil invertebrate biomass and play an essential role in many natural cycles (Edwards and Bohlen 1996; Lee 1985). Earthworms may therefore be considered effective biological indicators of soil quality (Blair et al.

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1996). In turf ecosystems, including lawns, earthworms are positively associated with many favorable soil characteristics including low amounts of thatch, low bulk density, high water infiltration and percolation rates, high organic matter concentrations, deep rooting, low plant wilting proneness, low plant disease severity, and uniform dispersion of microbes (Christians 1998; Turgeon 1999).

Professional lawn-care services utilize many types of chemicals including fertilizers, fungicides, herbicides, insecticides, and plant growth regulators. Standard sources (e.g., Christians 1998; Vengris and Torello 1982) recommend as many as nine fertilizer applications and six herbicide applications annually, with insecticides applied as needed. Even when used at labeled rates some registered lawn-care chemicals are highly toxic to earthworms and other soil invertebrates, while others are less toxic but still cause chronic effects and significant mortality (Potter 1994; Potter et al. 1990). To date, much research dealing with the toxicity of lawn care chemicals has been performed in laboratories or under controlled conditions at research facilities. Edwards and Bohlen (1996, Appendix A), for example, provided a summary of the specific effects of about 200 individual chemicals on earthworms, and Potter et al. (1990, 1994) described the effects of 40 commonly used pesticides and plant growth regulators on earthworms in turf.

Unlike controlled research plots, real lawns are simultaneously subject to many types of chemicals and to other types of stress such as dessication and compaction. In our study, we sampled earthworms in real lawns and compared populations in high-maintenance lawns (maintained by professional lawn-care companies using synthetic chemicals) to those in low-maintenance lawns (maintained by individual owners by mowing but without using chemicals).

## MATERIALS AND METHODS

### Study Area

Our study was conducted in Lexington, Fayette County, the second largest city in Kentucky. Lexington, founded about 1779, presently occupies about 12,000 ha and has a population of ca. 242,000. The landscape is a

rolling karst plain underlain by phosphatic Lexington Limestone of Ordovician age. Although the study area is largely urbanized, native soils are mapped as the Maury-McAfee Association (Sims et al. 1968). About 85% of the map unit is deep and moderately deep, well-drained soils in residuum on uplands, with silt loam A horizons, silty clay loam B horizons, and clay C horizons (Alfisols and Ultisols in the USDA taxonomy); the remaining 15% is silt loam soils in alluvium in sinkholes and drains (Mollisols and Inceptisols).

The climate is temperate, humid, and continental. Mean annual temperature and precipitation, respectively, are 13°C and 1148 mm. Weather during the 5-day sampling period of 6 Apr–10 Apr 1998 was normal, with maximum and minimum daily temperatures of 18°C and 7°C, and 30 mm of total precipitation. Soils were near optimal moisture content during the sampling period since there had been no extended dry periods and since rainfall during the preceding 12 months was ca. 8% above average (University of Kentucky Agricultural Weather Center 1999).

### Lawn Care Survey

A Lawn Care Survey was used to locate co-operators and to provide a pool of potential experimental lawns. The survey asked whether the lawn was maintained using lawn-care chemicals (herbicides, fertilizers, etc.), and, if so, if the lawn was maintained by a professional lawn-care service. In addition, the survey identified age of the house, length of time the lawn was maintained under the current maintenance regimen, and other factors that could influence our research. A total of 340 survey forms was distributed through the administrative offices of the Math, Science & Technology Center and the University of Kentucky. Two hundred and four completed survey forms were returned. To minimize variability in maintenance regimen, we first eliminated all lawns that were maintained by owners themselves using lawn-care chemicals. We also eliminated lawns with underground irrigation systems and fenced-in pets. We then carried out a stratified-random sampling of the remaining lawns to select 15 high-maintenance and 15 low-maintenance lawns with about equal distributions in terms of geographic location within Lexington, age of



Table 1. Characteristics of earthworm populations and of mature and immature earthworm specimens in lawns maintained by individual owners without the use of lawn care chemicals (low-maintenance) and in lawns maintained by professional lawn care services (high-maintenance) in Lexington, Kentucky. Within a row, different letters indicate that the characteristic differed between maintenance types at the indicated level of significance ( $P$ ) using Mann-Whitney tests;  $n = 15$  of each type lawn.

Earthworm characteristic	Low-maintenance	High-maintenance	<i>P</i>
	Median value		
Number (no/sample frame)	3.7 b	6.0 a	0.03
Total dry mass (mg/sample frame)	320 a	187 b	0.02
Individual dry mass, mature (mg/individual)	130 a	68 b	0.007
Individual dry mass, immature (mg/individual)	43 a	19 b	<0.001

house, and length of time the lawn was managed under the current maintenance regimen. Thirty lawns were chosen since this was the number that could be sampled within 1 week, the maximum sampling period that we judged still short enough to avoid variability in weather or other factors that could cause earthworms to move deep into the soil or to aestivate. All experimental lawns were associated with single-family homes, were regularly mowed, and were less than 4000 m<sup>2</sup> in size.

#### Field and Laboratory Methods

Earthworms were collected during the period 6 Apr–10 Apr 1998. The sequence of sampling alternated between high-maintenance and low-maintenance lawns, with randomized order within each type of lawn. On each lawn, a single random point was chosen away from the influence of structures, walkways, trees, and plant beds. Grass was clipped and thatch removed from a small area surrounding the point, and a square 0.1-m<sup>2</sup> frame was located on the prepared area. Next, 4 liters of 0.05% formalin was poured into the soil within the frame. This chemical acts as an irritant that causes many types of earthworms to emerge from soil (Lee 1985) and is considered the single best method for routine monitoring of lumbricids (Blair et al. 1996). We collected all earthworms flushed from the soil within the frame during a 10-minute period immediately after adding the formalin solution. We used the dry mass of these earthworms as the experimental variable to test for difference in earthworm abundance in high- and low-maintenance lawns.

Specimens were returned to the laboratory and anaesthetized in 10% EtOH. Each specimen was identified to genus (immature) or

species (mature); anaesthetized length and average diameter were measured.

#### Calculations and Statistical Analysis

Dry mass of each specimen was calculated by using anaesthetized length and diameter to calculate volume, assuming the anaesthetized shape to be a right circular cylinder. Volume was then converted to dry mass using Edward's (1967) average values for density (1.064 g/cm<sup>3</sup>) and percentage dry matter (26%) of lumbricids.

Non-parametric Mann-Whitney tests were used to compare high-maintenance versus low-maintenance lawns in terms of the number and total dry mass of earthworms collected per plot and the dry mass of individual earthworms. This was done for all specimens together and for immature and mature specimens separately. Spearman's correlations were used to examine relationships between number and mass of earthworms, age of house, and length of management regimen. Results of all procedures were evaluated at the 0.05 level of significance. All statistical analyses were done with the SPSS computer program (SPSS 1997).

#### RESULTS

There was no significant difference between high-maintenance and low-maintenance lawns in the age of the house ( $P = 0.68$ ) or the length of time the lawn was managed under the current maintenance regimen ( $P = 0.08$ ). Median values for house age and length of maintenance, respectively, were 26 yr and 12 yr. The genera *Lumbricus*, *Aporrectodea*, and *Eiseniella* were recorded, with *Lumbricus* dominant in both types of lawns, accounting for 89% of the specimens collected. There was



no significant difference ( $P = 0.19$ ) between the two types of lawns in the proportion of specimens that were immature; the median value was 72%. Maximum earthworm lengths were 60 and 65 mm, respectively, in high-maintenance and low-maintenance lawns; large specimens of *L. terrestris* were not collected. Significantly more earthworms per unit area were collected from high-maintenance versus low-maintenance lawns (Table 1). Conversely, low-maintenance lawns had significantly greater earthworm dry mass per unit area and dry mass per individual earthworm (Table 1). Neither number nor dry mass of earthworms was significantly correlated with house age or length of management.

## DISCUSSION

The conclusion I formed was that the lawn earthworm is a starved earthworm. . . . The lawn earthworms were much smaller and were not nearly so vigorous in their movements. . . . The wonder was that worms should be found living in such numbers in the lawn soil in these somewhat unnatural conditions.

W. H. Hudson 1919, p. 345–346

Our study of high- and low-maintenance lawns supports the conclusion reached 80 years ago by W. H. Hudson (see quote above). The mass of individual earthworms from low-maintenance lawns was similar to the normal range reported by Lee for Lumbricidae (1985, Table 7) whereas the mass of individual earthworms from high-maintenance lawns was about half normal. Although it is difficult to estimate earthworm densities using the formalin extraction technique (Lee 1985; Edwards and Bohlen 1996), densities calculated from our Table 1 of ca. 80 and 120 individuals/m<sup>2</sup>, respectively, for low-maintenance and high-maintenance lawns are far below the 100 to >2000 individuals/m<sup>2</sup> recorded by Lee for Lumbricidae in temperate pastures (1985, Table 7). We interpret this as indicating that earthworm growth in our study was more likely stunted by the lawn-care chemicals used in high-maintenance lawns than by competitive interactions or overcrowding. Other research also supports this conclusion by showing that some lawn and agricultural chemicals may adversely affect non-target organisms including earthworms and other soil invertebrates (Potter 1994; Potter et al. 1990). In addition to

direct toxicity, such adverse effects may be sublethal and chronic, resulting in slow growth or weight loss (Edwards and Bohlen 1996). Since low-maintenance and high-maintenance lawns were similar in terms of house age and length of time each maintenance regimen had been in use, the direct effect of maintenance regimen was not confounded by residues from past practices or by unequal representation of old and new houses.

Invertebrates such as earthworms have both utilitarian and intrinsic value (Samways 1994). The utilitarian value of earthworms as one of the most important members of the soil fauna is widely recognized (Blair et al. 1996; Christians 1998; Edwards and Bohlen 1996; Lee 1985; Turgeon 1999). In addition, it may be argued that ethical consideration of the intrinsic value of all species requires that humans, at a minimum, avoid causing death or pain to other species when such avoidance has little or no effect on human welfare (Samways 1994). This means that chemicals harmful to earthworms and other soil invertebrates should be avoided unless essential to human survival, health, or opportunity for fulfillment.

Pimentel et al. (1992) reviewed a wide variety of literature and concluded that the environmental and public health costs of using pesticides were so high that even the contributions of pesticides to economic profitability of agriculture was questionable. Furthermore, it was concluded that in agriculture it was possible to reduce pesticide usage by one-half with only minor effects on food prices since, in large part, pesticide use was driven by high cosmetic standards rather than by nutritional standards or plant health requirements (Pimentel et al. 1991). This suggests that lawn maintenance, which is clearly less essential to humans than agriculture, should be based on safer and less damaging practices than presently used. In particular, utilization of organic methods for restoring and maintaining soil fertility and health, or of native flora or other plant types that can be more naturally and easily maintained than turf, may be reasonable substitutes for the present system of chemical-based lawn care. Organic lawn care should become more popular with homeowners as information on organic practices (e.g., WSHU-FM and Duesing 1999) and commercial services (e.g., Bass Custom Landscapes, Inc.



1999) becomes available through the internet (e.g., WSHU-FM and Duesing 1999) and as recommendations on specific techniques become available through government organizations such as the USDA Cooperative Extension Service (e.g., Bruneau et al. 1997).

### ACKNOWLEDGMENTS

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## Influence of Topography on Local Distributions of *Plethodon cinereus* and *P. richmondi* (Plethodontidae) in Northern Kentucky and Southwestern Ohio

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### ABSTRACT

In northern Kentucky and southwestern Ohio, the more drought-tolerant ravine salamander, *Plethodon richmondi*, generally occupied regions with steeper slopes while the red-backed salamander, *P. cinereus*, was located in areas with less relief. On each of six slopes where their ranges overlapped, a relatively higher proportion of *P. richmondi* occurred on the upper area of the slope where drier conditions predominate, and a relatively higher proportion of *P. cinereus* existed on the lower portion of the slope where moist microhabitats are more common. Within the range of *P. cinereus*, four isolated *P. richmondi* populations coexisted with *P. cinereus* on dry nose slopes. The local distributions of *P. cinereus* and *P. richmondi* are related to topographic features affecting soil moisture content.

### INTRODUCTION

The geographic ranges of the red-backed salamander, *Plethodon cinereus*, and the ravine salamander, *P. richmondi*, overlap in eight states (Petranka 1998). However, the species usually are not syntopic in their areas of overlap (Highton 1972; Minton 1972; Pfingsten 1989a). Pfingsten (1989b), for example, reported that *P. richmondi* replaces *P. cinereus* in three eastern Ohio counties that contain some of the steepest slopes in the state. The purpose of my study was to investigate the influence of topographic features on the local distributions of the two salamanders in northern Kentucky and southwestern Ohio.

### METHODS

From 1991 through 1997, I conducted spring and autumn searches for the presence of *P. cinereus* and *P. richmondi* (*P. electromorphus* sp. nov. according to Highton 1999) at 202 locations in Boone, Kenton, and Campbell counties in Kentucky, and in Hamilton and Butler counties in Ohio. In 1998 and 1999, I returned to six valleyside locations of sympatry and surveyed the salamander population at each as I ascended along a series of switchbacks from the streambank at the bottom to the ridge at the top. Salamanders were found by overturning all manageable surface rocks and fallen logs encountered during my ascent. The animals and cover objects always were returned to their original positions.

### RESULTS AND DISCUSSION

Only *P. richmondi* populations were recorded from the rolling land in the unglaciated southern portion of the three Kentucky counties (Figure 1). In the glaciated area of Kentucky, exclusive *P. richmondi* populations were found only in the stream-dissected lands located near the Ohio River in northwestern Boone County and between the Licking and Ohio rivers in northeastern Kenton County and northern Campbell County. In glaciated southwestern Ohio, exclusive *P. richmondi* populations in Hamilton County were found only in the stream-dissected areas between the Great Miami and Ohio rivers and between the Little Miami and Ohio rivers. *Plethodon richmondi* populations in Butler County were located along valley slopes of the Great Miami River and its tributaries.

In total, exclusive populations of *P. richmondi* occurred at 63 sites. Exclusive populations of *P. cinereus* occurred at 111 sites, all located within the glaciated areas of Kentucky and Ohio that are less dissected. In general, *P. richmondi* occupied regions with steeper slopes while *P. cinereus* was located in areas with less relief.

Pfingsten (1989b) characterized *P. richmondi* as a more drought-resistant species, and Thurow (1968) suggested that *P. richmondi* replaces *P. cinereus* at sites that are slightly drier in summer. The contrasting ranges of the two species in northern Kentucky and southwest-



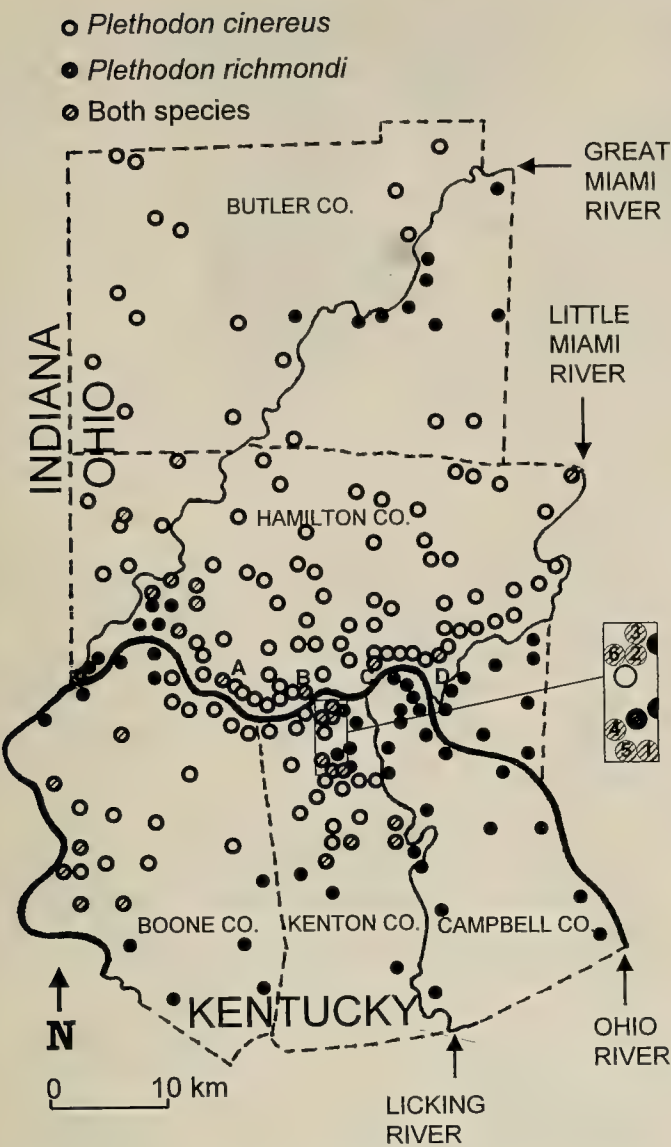


Figure 1. Occurrence of *P. cinereus* and *P. richmondi* in northern Kentucky and southwestern Ohio. The fully forested sympatric sites are labeled 1 to 6. The sympatric sites on nose slopes are labeled A to D.

ern Ohio perhaps are due to soil moisture differences caused by varying slope drainage conditions. *Plethodon richmondi* may be able to tolerate the well-drained, drier conditions

found in areas of greater relief where *P. cinereus* is absent. The two species were found together at 28 sites, including 24 valley slopes where their local ranges overlapped. To ascertain if *P. cinereus* and *P. richmondi* populations exhibited different distribution patterns in these areas of overlap, I surveyed the sympatric salamander communities on the six slopes that were fully forested (Figure 1, sites 1–6).

The number of animals captured at the sites ranged from 28 to 184, reflecting differences in slope length and in the number of cover sites provided by surface litter (Table 1). On five of the six slopes there was a significant difference between the distribution of the two species (Kolmogorov-Smirnov Test,  $P < 0.05$ ). On all six slopes the ratio of *P. cinereus* to *P. richmondi* was greater among the first third of the salamanders counted than among the remaining animals recorded further uphill (sign test,  $P < 0.05$ ).

The slope sampling data indicate a relationship between salamander distribution and soil moisture conditions related to topographic features. Compared to the upper two-thirds of a forested valley slope, the soil on the lower third of the slope holds the greatest moisture (Thomas and Anderson 1993). The soil on the lower portion is deeper, receives more runoff, and is less exposed to factors causing evaporation. On each valley slope where sympatric *P. cinereus* and *P. richmondi* were surveyed, a relatively higher proportion of *P. cinereus* occurred on the lower portion of the slope where moist microhabitats are more common, while a relatively higher proportion of *P. richmondi* existed on the upper area of the slope where drier conditions predominate.

Finally, at four of the 28 sites of sympatry

Table 1. Significance and comparisons of different distributions of *P. cinereus* and *P. richmondi* in uphill counts on six valley slopes in Kenton County, Kentucky. Significance of different distributions is based on the Kolmogorov-Smirnov Test; n.s. is non-significant.

Slope	Date	P	Total count	<i>P. cinereus</i> : <i>P. richmondi</i> ratio	
				First third of count	Remainder of count
1	30 Mar 98	n.s.	28	9:1 (9.0/1)	10:8 (1.3/1)
2	18 Apr 98	0.007	70	19:5 (3.8/1)	16:30 (0.5/1)
3	25 Apr 98	0.001	65	19:3 (6.3/1)	19:24 (0.8/1)
4	29 Apr 98	0.032	184	58:4 (14.5/1)	95:27 (3.5/1)
5	2 Apr 99	0.033	132	37:7 (5.3/1)	66:22 (3.0/1)
6	25 Apr 99	0.049	83	26:2 (13.0/1)	40:15 (2.7/1)



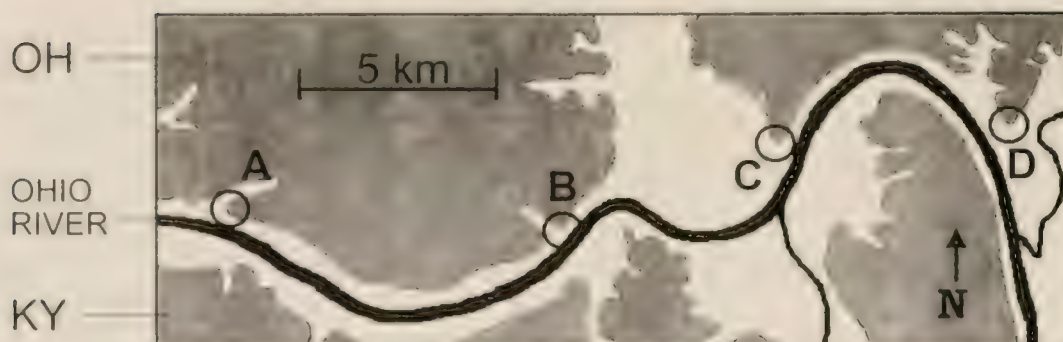


Figure 2. Map of nose slopes A to D above the Ohio River in Hamilton County, Ohio. The shaded areas are 200 to 285 m above sea level.

the *P. richmondi* population was isolated from any adjoining population of the same species (Figure 1, sites A-D). All these sites were located on nose slopes, named for their resemblance to the convexity of a human nose (Figure 2). *Plethodon richmondi* probably was able to coexist with *P. cinereus* on these nose slopes because varying soil moisture alternately favored one species over the other.

A nose slope is formed where the side slope of a river valley is incised by a tributary leading to the river. The resulting promontory has the river side slope along one edge and the tributary side slope along the other. The two side slopes are connected at the tip of the promontory by the nose slope.

Downhill flows of storm runoff on a side slope are roughly parallel (Hole and Campbell 1985). In contrast, water disperses as it flows downhill on a convex nose slope, causing drier soil conditions on a nose slope than on the adjacent side slopes. The relative dryness of a nose slope also is due in part to its greater exposure to desiccating air currents at the end of the promontory (Thomas and Anderson 1993).

Evaporation caused by sunlight also may contribute to the aridity of nose slopes (Buol et al. 1989). In the Northern Hemisphere, south- and west-facing slopes are drier and north- and east-facing slopes are moister. The west aspect of nose slope A and the south aspect of nose slopes B-D probably help to promote the frequent reduction of soil moisture content to levels that permit *P. richmondi* to coexist with *P. cinereus*.

Substrate moisture differences caused by variations in the landscape are responsible for the separation of *P. cinereus* from the more

drought-tolerant *P. shenandoah* (Jaeger 1971), *P. hoffmani* (Highton 1972), and *P. wehrlei* (Pauley 1978). Local distributions of *P. cinereus* and *P. richmondi* probably also are related to soil moisture levels that are governed by topographic features.

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# Comparison of Macroinvertebrate Communities of Two Intermittent Streams with Different Disturbance Histories in Letcher County, Kentucky

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## ABSTRACT

Small headwater streams receive little attention with regard to land management and regulatory policy, yet they greatly contribute to regional biodiversity and ecosystem function of their receiving streams. To ascertain differences associated with past logging and mining disturbances, I surveyed the macroinvertebrate communities of two first-order streams in the Eastern Coalfield Region (Letcher County, Kentucky) in spring, summer, fall, and winter 1998–1999. The reference stream drains old-growth forest in Lilley Cornett Woods; the disturbed stream lies in an adjacent hollow that was logged ca. 1940s and contour-mined ca. 1975. Benthic macroinvertebrates were collected using both quantitative and qualitative techniques at one site in each watershed. Results showed that the reference stream scored higher in measures of taxon richness, EPT richness, density, and diversity ( $H'$ ) but did not differ greatly in the pollution tolerance index (mHBI) or functional feeding group organization. Seasonal differences in taxon richness, EPT richness, density, and diversity within individual streams were noted and were attributed to life history phenologies of the resident taxa. Both streams had dominant taxa in common in spring (e.g., *Paraleptophlebia*, *Epeorus*), summer (e.g., *Leuctra*, *Paraleptophlebia*), fall (e.g., *Diplectrona*, *Paraleptophlebia*), and winter (e.g., *Ephemerella*, *Epeorus*), but wide variation in relative abundances were observed for most species. These results provide a reference for future comparisons of macroinvertebrate community structure in these and other small streams in the region.

## INTRODUCTION

Streams draining old-growth forests offer valuable reference information on stream ecosystem structure and function. In Kentucky, only remnants of old growth forest exist, but they still provide a valuable source of baseline ecological data. Stream systems degraded by anthropogenic activities such as agriculture, logging, mining, and urbanization are widespread in Kentucky (Kentucky Division of Water 1996). In the eastern coal field region of the state, many small streams have been directly impacted by surface mining and recent logging. Therefore, it is important to document those biological communities in the remaining undisturbed streams. Since the turn of the century, aquatic organisms have been used extensively in water quality monitoring and impact assessment (Cairns and Pratt 1993). In addition to their use in biological monitoring and assessment, aquatic invertebrates are extremely important in stream ecosystem processes (Cummins 1974). Furthermore, general inventories of invertebrate taxonomic groups are needed to help document and explain patterns of biodiversity in Kentucky.

Lilley Cornett Woods (LCW) has served as an Appalachian ecological research station since 1969. It has been the focus of numerous environmental studies including hydrogeology (Conrad 1983); vegetation (Martin 1975; Martin and Sheperd 1973; Muller 1982; Sole et al. 1983; and others); mammals (Barels 1985); birds (Hudson 1972; Schwierjohann and Elliott unpub. data); and reptiles and amphibians (Cupp and Towels 1983; Towels unpubl. data). My study compared macroinvertebrate community structure in an undisturbed, first-order stream in Big Everidge Hollow (BEH), an old-growth forested watershed in LCW, with an adjacent first-order stream in Poll Branch Hollow (PBH) that had been previously logged and contour mined. Objectives were (1) to estimate community composition and structure in an exceptional, first-order stream in the Eastern Coalfield region, (2) to compare fauna occurring in an “old-growth” stream and a “second-growth” stream having previous coal mining disturbances, and (3) to determine seasonal differences in community structure in these streams. This was the first extensive survey of benthic macroinvertebrates in BEH and



Table 1. Selected physico-chemical characteristics of Poll Branch Hollow and Big Everidge Hollow, Letcher County, Kentucky. Parameters marked with an asterisk (\*) were measured on 12 Dec 1998.

	Big Everidge Hollow	Poll Branch Hollow
Watershed area (ha)	~60	~90
Site elevation (m)	~340	~340
Aspect	East	East
Channel		
Gradient	7%	6%
Length (m)	800	900
Width (m)	0.5–2.5	0.5–2.5
Depth (cm)	2–45	2–25
Canopy	Full	Full
Temperature °C	7.3–17.8	6.7–17.6
Conductivity (µmho)	52–72	126–161
pH (S.U.)	7.4–7.6	7.1–7.7
Total hardness (mg/liter)*	26.5	58.2
Sulfate (mg/liter)*	12.3	35.1
Aluminum (mg/liter)*	0.05	0.43
Iron (mg/liter)*	0.06	0.89
Manganese (mg/liter)*	0.001	0.088

should provide a foundation for future comparisons.

### STUDY AREA

LCW is a 222 ha natural preserve north of Pine Mountain in southeastern Kentucky in the Cumberland Plateau section of the Appalachian Plateau physiographic province. The preserve lies within the Tilford and Roxanna USGS 7.5 minute quadrangles. Both BEH and PBH are east-facing, first-order tributaries to Line Fork Creek, a fourth-order tributary to the North Fork Kentucky River. Both watersheds are underlain by interbedded sandstones, siltstones, shales, and coal. Physico-chemical attributes for the streams are shown in Table 1. The study streams may have periods of intermittency in late summer and early fall of dry years, but they may remain perennial during wet summers (M. Brotsge, LCW, pers. comm., 12 Nov 1997). Mixed mesophytic forest made up the riparian corridor in both streams and was dominated by *Acer rubrum*, *Aesculus octandra*, *Fagus grandifolia*, *Liriodendron tulipifera*, and *Tsuga canadensis* (in alphabetical order). The upland slope assemblage in BEH consisted of old-growth stands of *Acer saccharum*, *Carya* spp., *Fagus grandifolia*, *Liriodendron tulipifera*, and *Quercus alba*; and ridge top forest was dominated by old-growth *Carya* spp., *Quercus montana*, and

*Quercus coccinea*. The upland forest in PBH is second-growth and generally consists of the same species typical of BEH (Muller 1982).

A single 100 m sampling reach was selected in the lower portion of each watershed. Both channels are tightly constrained, bedrock-dominated streams having high gradient. Both fast and slow water bedrock habitat collectively comprised nearly 50% of the each sampling reach. However, each stream had a variety of geomorphological habitat units that included cobble-boulder riffles and pools, and bedrock glides and trench pools. Areas of fine gravel, sand, and silt were limited to pool areas and the margins of slow riffles. However, upstream of the sample reach in PBH, access roads and a sediment retention dam were constructed for mining operations in the mid 1970s. Since that time, the roads have been reforested but the dam has breached after filling with sediment. Despite natural reforestation, it was apparent that PBH experienced serious bank erosion upstream of the sample reach. It was also evident that interstitial substrates were burdened with excessive silt fines, and the degree of embeddedness was greater in riffle habitats compared to BEH. With regard to stream vertebrates, desmognathine salamanders were commonly observed in both streams. The headwater position of the streams preclude the establishment of a diverse fish community; the only species observed was the creek chub, *Semotilus atromaculatus*.

### METHODS

#### Sample Collection

I collected macroinvertebrates using both quantitative and qualitative techniques once in spring (16 Apr 1998), early summer (24 Jun 1998), late fall (12 Dec 1998), and winter (9 Feb 1999). Quantitative data were taken from four replicate Surber samples (0.09 m<sup>2</sup>, 750 µm mesh) stratified along a longitudinal transect within the thalweg (i.e., path of deepest thread of water) of a cobble-pebble riffle. This habitat was targeted to ensure the highest species richness and abundance of macroinvertebrates (Brown and Brussock 1991; Feminella 1996). Samples were elutriated with a wash basin and a 600 µm (U.S. No. 30) sieve and preserved in pint jars containing 95% ethyl al-



cohol. An effort was made to remove much of the leaf debris and many of the larger stones collected in the Surber samplers prior to sieving. This was accomplished by inspecting and washing individual leaves and stones in the wash basin. Qualitative species collections were gathered from multiple-habitat (i.e., woody debris, leaf packs, moss, large slab rocks, etc.) hand picking and dipnet (D-frame,  $800 \times 900 \mu\text{m}$  mesh) sampling for ca. 30 min. (except 45 m. for winter sample), and specimens were preserved in 70% ethyl alcohol. In the laboratory, entire samples were picked in an enamel pan without magnification until no more invertebrates were found and then briefly viewed at  $20\times$  under a dissecting microscope to search for smaller, cryptic forms. Temperature, dissolved oxygen, pH, and conductivity were measured on each sampling date with a portable Hydrolab meter. Water samples were collected from each stream on 12 Dec 1998, and chemical variables were analyzed by the Kentucky Department for Environmental Services, Frankfort, Kentucky.

#### Data Analysis

Various community metrics were calculated in an effort to describe macroinvertebrate community structure between sites and seasons. Measures of richness and abundance were determined for all seasons. The Ephemeroptera, Plecoptera, and Trichoptera (EPT) index (a measure of the richness of those generally pollution-sensitive insect orders) was also calculated. For comparison, the Shannon diversity index ( $H'$ , Shannon 1948) was determined using the base<sub>e</sub> logarithm. Overall biotic health of the stream was measured with the modified Hilsenhoff Biotic Index (mHBI), or North Carolina Biotic Index (NCBI, Lenat 1993), a weighted index based on individual pollution tolerance values and species proportions in the community. Tolerance values range from 0 (intolerant) to 10 (tolerant). Among sites and seasons, Mann Whitney U-tests were used to test for significant differences in means of taxon and EPT richness, abundance, and diversity at  $P < 0.05$ . The relative abundance of functional feeding groups was also calculated. Functional feeding group assignments followed Merritt and Cummins (1996) and Thorp and Covich (1993). Differences in overall taxonomic composition were

illustrated with the % community similarity index (Sokal and Rohlf 1973), and the Ten Dominants in Common metric ( $\text{DIC}_{10}$ ). To demonstrate differences in community composition, % community similarity dendrograms were constructed using hierarchical (UPGMA) cluster analysis (Romesburg 1990).

## RESULTS

### Water Chemistry

Although a detailed investigation of water quality was not conducted, notable differences were found between sites (Table 1). Compared to BEH, elevated conductivity, various metals, sulfate, and total hardness were the most distinctive parameters found in PBH. In some instances, values in PBH were several times higher than those in BEH. The ranges of temperature and pH between sites were nearly identical on each sampling occasion (Table 1).

### Community Composition

A total of 118 taxa representing 14 orders and 45 families was collected in both streams combined during all sampling efforts (Appendix A). The most taxa were recorded in BEH (106 taxa) compared to PBH (83). In BEH, the insect order Diptera had the most taxa (37), followed by Trichoptera (21), Plecoptera (15), and Ephemeroptera (15) in all seasons combined. By contrast, in PBH Diptera had the most taxa (31), followed by Ephemeroptera (17), Trichoptera (13), and Plecoptera (12). Seasonal trends in total taxon richness are shown in Appendix A.

Comparisons of site and seasonal trends in taxon richness, EPT richness, density, diversity, and the mHBI for both streams are shown in Table 2. Within individual streams, mean taxon richness, EPT richness, and densities were significantly higher in spring and winter samples ( $P < 0.05$ ), suggesting a strong seasonal component to community structure. Seasonal differences in Shannon diversity were negligible in both streams. With regard to pollution tolerance (mHBI), all streams consistently scored in the excellent range ( $\text{mHBI} < 3.3$ ). Between sites, taxon richness was higher in BEH throughout all seasons, although the difference during the winter was not significant (Table 2). Although mean EPT richness in BEH was greater among all sea-



Table 2. Mean ( $\pm 95\%CI$ ,  $n = 4$ ) benthic macroinvertebrate richness, combined number of taxa within the insect orders Ephemeroptera, Plecoptera, and Trichoptera (EPT) richness, density  $m^{-2}$ , and Shannon Diversity ( $H'$ ) among Surber samples ( $0.09\ m^2$ ) taken from Big Everidge Hollow (BEH) and Poll Branch Hollow (PBH), Letcher County, Kentucky. The modified Hilsenhoff Biotic Index (mHBI), % community similarity, and number of the top 10 dominant taxa in common ( $DIC_{10}$ ) were calculated from the four composited Surber samples. Spring (Sp), summer (Su), fall (F), and winter (W) correspond to mid-April, late-June, early December, and early February, respectively. An asterisk (\*) indicates significantly higher values at the  $P < 0.05$  level.

	Sp		Su		F		W	
	BEH	PBH	BEH	PBH	BEH	PBH	BEH	PBH
Taxon Richness	32.0 $\pm$ 5.1*	19.2 $\pm$ 6.1	20.2 $\pm$ 5.4*	11.7 $\pm$ 0.5	24.0 $\pm$ 7.4*	15 $\pm$ 2.4	25.3 $\pm$ 3.2	24.5 $\pm$ 4.5
EPT Richness	17.5 $\pm$ 1.9	13.2 $\pm$ 3.2	11.0 $\pm$ 2.9	7.2 $\pm$ 2.5	12.8 $\pm$ 2.9	10.2 $\pm$ 0.2	16 $\pm$ 2.1	14.3 $\pm$ 3.0
Density $m^{-2}$	1936 $\pm$ 871*	811 $\pm$ 370	844 $\pm$ 449*	317 $\pm$ 52	572 $\pm$ 232	381 $\pm$ 117	1433 $\pm$ 58	1330 $\pm$ 298
$H'$ Diversity	2.7 $\pm$ 0.1	2.5 $\pm$ 0.2	2.6 $\pm$ 0.3*	2.2 $\pm$ 0.1	2.9 $\pm$ 0.3*	2.3 $\pm$ 0.2	2.7 $\pm$ 0.2	2.6 $\pm$ 0.1
mHBI	2.49	2.60	1.93	2.79	3.27	2.49	1.81	1.97
% Similarity								
$DIC_{10}$	47	8	41	4	43	67	6	6

sons, values were not significantly different ( $P < 0.05$ ). Densities of macroinvertebrates were consistently higher in BEH, but only spring and summer collections were significant. Shannon diversity was also higher in BEH among seasons but was not significant in spring. Relatively low percent community similarity was seen in all seasons, with the exception of winter where the communities were 67% similar. The lowest similarity was found in summer (40%). Comparatively, the  $DIC_{10}$  metric portrayed the greatest similarity in spring and winter. A dendrogram illustrating community similarity among sites is shown in Figure 1.

A few individual taxa consistently showed higher abundances in quantitative samples among sites and seasons. Table 3 lists the top five taxa occurring at each site among seasons. The leptophlebiid mayfly *Paraleptophlebia* was a dominant taxon during all seasons in BEH; it was dominant in PBH three of the four seasons. For PBH, the heptageniid mayfly *Epeorus* and the hydropsychid caddisfly *Diplectrona* were dominant taxa during three of the four seasons; in BEH these taxa were abundant in two and three seasons, respectively. The stonefly *Leuctra* was another notable taxon, being common in both streams two of four seasons.

### Functional Feeding Groups

Functional feeding groups were very similar among sites and seasons (Figure 2). The relative proportion of shredders (detritivores adapted to feed upon leaf and woody debris, or coarse particulate organic matter (CPOM,  $>1\ mm$ )) and invertebrate predators were the most consistent between sites and seasons. Collectors (detritivores feeding primarily upon fine particulate organic matter (FPOM,  $0.5\text{--}1\ mm$ ) deposited either on the substrate surface or within the interstices, or suspended in the water column) and scrapers (grazing herbivores feeding upon periphyton and associated material) were more variable in their abundances among sites and seasons. Collectors were the most abundant group in both streams in all seasons, ranging from 33% to 56% in BEH and 42% to 50% in PBH. Scrapers were the second most abundant group and reached their greatest abundance in winter with lowest proportions observed in summer



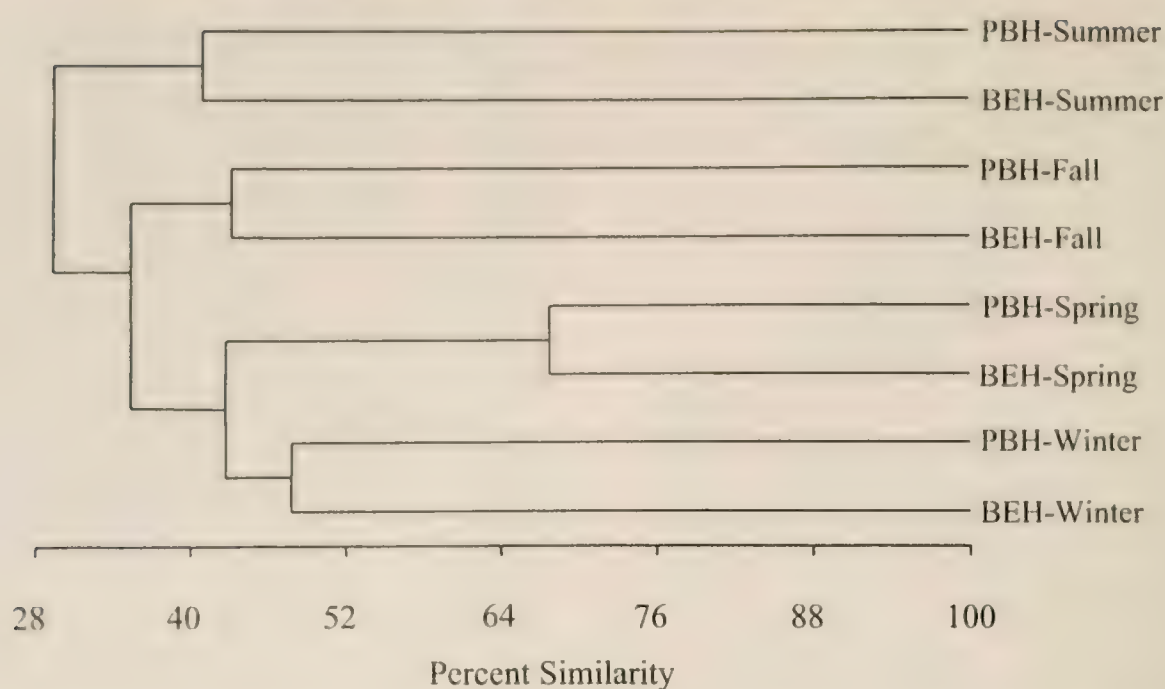


Figure 1 Dendrogram showing percent similarity between Poll Branch Hollow (PBH) and Big Everidge Hollow (BEH), Letcher County, Kentucky among seasons 1998-1999.

and fall. Relative proportions ranged from 17% to 33% in BEH and 13% to 32% in PBH. Shredders represented a smaller proportion at both sites, being most abundant in summer and least in winter. The relative abundance of shredders ranged from 12% to 20% in BEH and 9% to 23% in PBH. Similarly, predators were most abundant in summer and least in winter. Predator abundance ranged from 13% to 21% in BEH and 12% to 20% in PBH.

## DISCUSSION

### Community Composition

The invertebrate fauna in BEH and PBH consisted mainly of insect larvae typically associated with clean, high-gradient streams in the region. The dominant taxa found in this survey were immature insects that are considered to have univoltine life cycles (Brigham et al. 1982; Merritt and Cummins 1996; Stewart and Stark 1988; Wiggins 1996) and are known to inhabit both intermittent or perennial streams (Feminella 1996). With regard to life history, only a few species were thought to undergo semivoltinism in the study streams (e.g., *Acroneuria*, *Nigronia*, *Cordulegaster*, and *Stylomorphus*). The thermal regime exerts considerable influence on insect voltinism (Sweeney 1984) and in small, forested streams like BEH and PBH, cool annual temperatures may limit

the degree of multivoltinism, which is common in larger, warmer streams (Hynes 1970).

I considered overall taxon richness in both study areas to be fairly high, but values varied markedly among sampling season. By contrast, Harker et al. (1980) suggested that small pristine mountain streams in Kentucky may have reduced richness and diversity due to low nutrient or alkalinity values. Vannote and Sweeney (1980) also indicated that low flow, lower habitat diversity, and greater thermal constancy may limit invertebrate taxon richness in small streams. Seasonal differences in species richness are typical of small streams including BEH and PBH that are faced with periods of intermittency where life history adaptations determine species presence and absence. Total EPT richness, which is often highly correlated with taxon richness, was also high in each stream. Those EPT taxa, in general, represent a group of organisms that are intolerant of environmental stress including water pollution (Lenat 1988) and typically proliferate in clean mountain streams. In my study, BEH had 51 EPT taxa including 14 taxa not encountered in PBH, while PBH had 42 EPT taxa with 6 taxa not collected in BEH.

Based on my results, significant differences among richness in BEH and PBH cannot be entirely explained. Wagner and Benfield



Table 3. Seasonal differences in the percent composition of the 5 most abundant taxa occurring in composited Surber samples (n = 4) in Big Everidge Hollow (BEH) and Poll Branch Hollow (PBH), Letcher County, Kentucky during spring (mid-April), summer (late-June), fall (early-December), and winter (early-February).

	BEH	PBH
Spring	<i>Paraleptophlebia</i> (31) <i>Epeorus</i> (9) <i>Leuctra</i> (9) <i>Baetis intercalaris</i> (4) <i>Diplectrona</i> (4)	<i>Paraleptophlebia</i> (15) <i>Epeorus</i> (12) <i>Cinygmula</i> (11) <i>Amphinemura</i> (7) <i>Tanytarsus</i> (7)
Summer	<i>Stenonema meririvulatum</i> (17) <i>Leuctra</i> (15) <i>Paraleptophlebia</i> (9) <i>Leucrocuta</i> (7) <i>Sweltsa</i> (7)	<i>Leuctra</i> (21) <i>Ectopria</i> (10) <i>Paraleptophlebia</i> (10) <i>Diplectrona</i> (7) <i>Acroneuria</i> (7)
Fall	<i>Paraleptophlebia</i> (12) <i>Diplectrona</i> (10) <i>Ectopria</i> (7) <i>Ameletus</i> (5) <i>Parametriocnemus</i> (5)	<i>Diplectrona</i> (30) <i>Neophylax</i> (10) <i>Leuctra</i> (10) <i>Epeorus</i> (5) <i>Paraleptophlebia</i> (4)
Winter	<i>Epeorus</i> (24) <i>Ephemerella</i> (10) <i>Diplectrona</i> (8) <i>Baetis tricaudatus</i> (5) <i>Paraleptophlebia</i> (5)	<i>Ephemerella</i> (21) <i>Epeorus</i> (19) <i>Diplectrona</i> (10) <i>Sweltsa</i> (5) <i>Neophylax</i> (4)

(1998) found that old-growth forested streams in North Carolina had less biodiversity than catchments logged 85+ yr ago but greater biodiversity than watersheds logged 25 to 50 yr ago. Their data suggested that macroinvertebrate communities continue to be influenced by logging long after reforestation. In my study, it had been over 50 yr since PBH was clearcut, and 25 yr since it was strip-mined. Observations on substrate composition and microhabitat availability revealed that a moderate degree of embeddedness (~50%) was evident in PBH riffles and pools, indicating that interstitial spaces were more frequently clogged with fine sediment. This can result in decreased colonization area for benthic macroinvertebrates. Chronic bank erosion from upstream channel modification (i.e., past coal mining and associated instream settling pond and roads) contributes to substantial sedimentation and substrate embeddedness. Other factors contributing to lower taxon richness in PBH may be related to water chemistry (i.e., elevated metals and conductivity from coal mining) or disturbance history (i.e., logging and mining) and the possible extirpation of headwater species from PBH. With regard to water chemistry, the data found in my study are in very close agreement with those found

by Dyer (1982) who sampled mined and unmined streams having similar sized watersheds within the Line Fork drainage. In mined streams, he found elevated conductivity and metals in similar proportions as those found in PBH in my study. In undisturbed streams, his values were very similar to those I found in BEH.

Although many of the dominant taxa I found were common to both streams, differences in the less frequently collected taxa in BEH appeared to affect total and mean richness indices. For example, sensitive habitat specialists that are locally rare (e.g., the caddisflies *Molanna*, *Theliopsyche*, and *Goerita*), may be locally extirpated when exposed to severe or long-lasting disturbance events such as forest clear-cutting and mining. Drastic changes in the food resource base and increased water temperature are probably the driving factors behind faunistic change (Gurtz and Wallace 1984; Stone and Wallace 1998). However, severe sedimentation and embeddedness can cause substantial declines in insect abundance (Waters 1995) or perhaps eliminate rare species from particular stream reaches. Temporary extirpation is probable when severe disturbance events occur at the stream source where reinvasion by drifting organisms is dis-



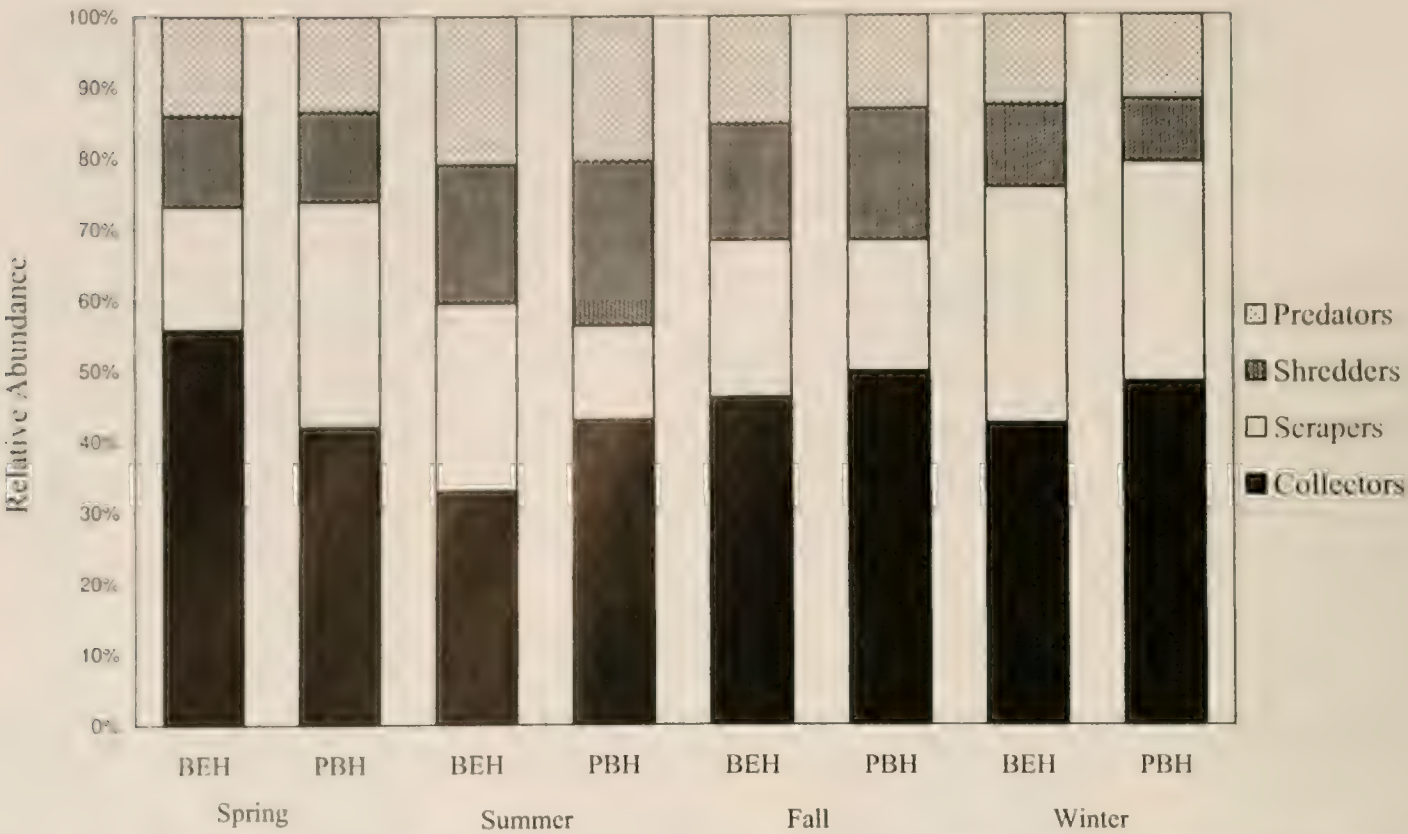


Figure 2. Relative Proportion of Functional Feeding Groups in Big Everidge Hollow (BEH) and Poll Branch Hollow (PBH), Letcher County, Kentucky among seasons 1998-1999.

rupted. Recolonization by adult dispersal is possible, although fauna restricted to headwater streams become somewhat isolated when the stream drains directly into larger streams (e.g., Line Fork Creek). In this case, colonization must occur by “over the mountain” adult dispersal which for some EPT taxa, is a “chance event” or may take considerable time (J. Morse, Clemson Univ., pers. comm., 6 Aug 1998).

Seasonal trends in estimated densities were consistently found among both study streams. Significant differences between the study streams are probably a result of previously mentioned embeddedness; however, water chemistry and disturbance history may also have an affect on macroinvertebrate abundance in PBH. Small headwater streams including BEH and PBH frequently undergo periods of intermittency during the summer months. This has a considerable impact on macroinvertebrate recruitment and overall relative densities from season to season as well as from year to year (Feminella 1996). Moreover, low nutrient or alkalinity levels associated with pristine streams may limit macroinvertebrate densities and secondary production on an annual basis.

Functional Feeding Groups

Although most benthic macroinvertebrates are considered omnivorous feeders, they can be categorized into “guilds,” or groups of organisms using a particular resource class. An analog to the trophic guild idea is the use of functional feeding groups based on an organism’s morpho-behavioral adaptations for food acquisition (Cummins 1973) rather than solely on the basis of what food is eaten. There is much debate on the overall utility of functional feeding group analysis since certain species are facultative feeders, or may show marked differences in food use among various life history stages (Allan 1995). However, the relative abundances of feeding guilds still provide useful information on the overall trophic organization and food resource dynamics of stream reaches.

The abundances of the various functional feeding groups did not vary greatly between streams. I expected shredders to be dominant in both streams based on observed inputs of leaf and wood debris into the stream from riparian vegetation. However, since quantitative sampling was conducted in riffles, the shredder component could be underestimated as



the niche of many shredders (e.g., *Eurylophella funeralis*, *Pycnopsyche* spp.) were found in pools that had accumulated more detritus than in riffles. Because benthic algal communities are limited by light in small streams, grazing herbivores (scrapers) are predicted to be less represented in forested headwater streams and to reach their greatest abundance in mid-order streams (Vannote et al. 1980). This was not the case in my study; scrapers were well represented in both streams. The unusually high abundance of scrapers was probably a relict of riffle sampling. Collectors further contribute to detritus breakdown in all aquatic systems and their abundance gives a general indication of how much organic matter is stored within a stream reach or specific habitat. Collectors outnumbered other feeding groups in both study streams in each season, indicating fine detritus as the dominant food-particle size. Unlike the seasonally available food resources used by shredders and scrapers, collectors are not generally limited by seasonality since FPOM transport and deposition is flow dependent and may be affected by biological processing rates (Webster 1983). The relative abundance of predators is generally predicted to remain constant both spatially and temporally along the stream continuum (Vannote et al. 1980). The proportion of this trophic group in each stream appeared to follow this prediction with respect to seasonality and the headwater position of the stream.

#### Diversity and the Biotic Index

The diversity and biotic indices implied exceptional water quality in BEH and PBH. Very little seasonal differences were observed in these community-level attributes, suggesting temporal stability in the communities. The low biotic index values showed that pollution-intolerant species dominated both streams. Although the mHBI responds primarily to organic or other toxic pollution, it is less sensitive to sedimentation unless the problem is both severe and chronic (pers. obs.). Stone and Wallace (1998) reported that after 16 years of forest succession, mHBI (NCBI) values were not significantly different between a reference stream and one disturbed by clear-cut logging. Since BEH is undisturbed, the presence of several tolerant taxa ( $TV > 7.0$ )

may suggest that these taxa are “colonists” or “ecological generalists” and that the tolerance values demonstrate euryoecic characteristics. However, the proportion of invertebrates having high tolerance values in BEH was low, implying that these taxa were sporadic colonizers of the stream.

Although the % similarity metric strongly suggested different communities among sites (except in winter), assemblages generally contained many overlapping taxa. Despite low % similarity between sites, it appeared that seasonality was the main factor affecting community composition as indicated by the cluster analyses. The lower degree of community similarity between seasons was anticipated since many of the univoltine species can be temporally absent from the community depending on individual life histories. Egg diapause is common in headwater stream invertebrates, and many species remain within hyporheic habitats for extended periods (Williams 1987) and thus appear to be seasonally absent. These seasonal influences appeared to outweigh differences possibly associated with disturbance history.

#### CONCLUSION

In this study I found that the macroinvertebrate communities in BEH and PBH represented rich and diverse faunas dominated by aquatic insects that are considered to be highly sensitive to anthropogenic disturbances. However, significant differences and dissimilarities in community structure were observed. The reduced number of species and density of individuals in PBH were possibly related to microhabitat differences among riffle substrates or water chemistry. Riffle embeddedness was probably the most notable microhabitat feature limiting macroinvertebrates in PBH. Both logging and mining through stream channels have the potential to affect downstream habitats by intensified erosion and sedimentation. The fact that BEH could consistently support more species and individuals sheds light on the importance of undisturbed watersheds for sustaining invertebrate biodiversity and production. Despite these pair-wise differences, seasonality was the driving force behind overall community structure within respective streams. Functionally, the study streams displayed similar feeding guild abundances, in-



dicating no differences in the food-energy base. No exceptionally rare species were collected; however, one stonefly genus (*Yugus*) is reported for the first time in Kentucky; infrequently collected caddisflies like *Molanna* and *Theliopsyche* were found in BEH. Based on my observations of other first-order streams in southeastern Kentucky, the present study revealed that the stream draining BEH did not necessarily harbor a unique stream assemblage but nevertheless characterized a sensitive and diverse community that has adapted to relatively continuous, natural ecosystem processes and may be suggestive of the prehistoric norm. However, because of the intermittent nature of BEH and PBH, extreme variation in community structure is possible from year to year. This necessitates the need for continued biological monitoring in these streams. Furthermore, research on other aspects of stream ecosystem structure and function (e.g., nutrient cycling, organic matter retention and transport, primary and secondary production) in BEH and PBH would be valuable to our understanding of the dynamics of old-growth forested watersheds in Kentucky.

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Appendix A List of macroinvertebrates collected from Big Excridge Hollow (BEH) and Poll Branch Hollow (PBH) during spring (mid-April), summer (late June), fall (early-December), and winter (early-February) 1998-1999. Values are in percent composition. X denotes qualitative occurrence.

Class	Order	Family	Genus/Species	BEH				PBH			
				Sp	Su	F	W	Sp	Su	F	W
TURBELLARIA OLIGOCHEATA	Tricladia	Planariidae	planariid (immature)	0.9	0.3		X				
		Lumbriculidae	<i>Ecliptidrilus</i> sp.	0.7	0.7	4.9	1.0				
	Haplotaenia	Naididae	<i>Dero nivea</i>					3.4	5.3	1.5	X
BIVALVIA INSECTA	Veneroida Ephemeroptera	Tubificidae	<i>Pristina acquista</i>	0.1							
		Tubificidae	tubificid (immature)				X				
		Sphaeriidae	<i>Pisidium</i> sp.	0.1		0.5	X				
		Ameletidae	<i>Ameletus</i> sp.	2.5		5.3	3.1				
		Baetidae	<i>Acentrella ampla</i>	2.0							
			<i>Baetis tricaudatus</i>		4.6				0.9		
			<i>Baetis flavistriga</i>		0.3				3.5		
			<i>Baetis intercalaris</i>		4.0				1.8		
			<i>Baetis</i> sp. A	4.5							
			<i>Centropilum</i> sp.		0.3				1.8		
		Ephemeridae	<i>Ephemerella gutturala</i>	0.1		0.5	X		3.5		X
			<i>Ephemerella simulans</i>						0.9		
		Ephemerellidae	<i>Ephemerella</i> prob. <i>auravillii</i>	3.9		2.9	10.3			X	
			<i>Eurylophella funeralis</i>		0.3	0.5	X		0.9	0.7	21.5
			<i>Cinygmula subaequalis</i>	X			0.4			2.2	0.2
		Heptageniidae	<i>Epeorus</i> prob. <i>nanatus</i>	9.1		0.5	23.8			5.1	3.6
			<i>Leucrocuta</i> prob. <i>thetis</i>		6.9				1.8		19.0
			<i>Stenacron interpunctatum</i>	0.4		X	X			X	1.5
Plecoptera		Capniidae	<i>Stenonema mericulatum</i>	0.6	17.1	2.9	2.1		0.9		0.2
			<i>Stenonema</i> sp.								
			<i>Stenonema vicarium</i>								
		Leptophlebiidae	<i>Paraleptophlebia</i> prob. <i>ontario</i>	31.1	9.5	12.6	5.0		9.7	4.4	2.3
			<i>Allocapnia</i> sp.			X	X			X	
		Chloroperlidae	<i>Alloperla</i> sp.					2.4			
			<i>Haploperla</i> sp.	0.1							
			<i>Sweltsa</i> sp.	2.2	6.9	1.9	3.5		2.6	0.7	4.6
		Leuctridae	<i>Leuctra</i> sp.	8.6	15.1	4.9	5.0		21.9	10.2	2.7
		Nemouridae	<i>Amphimnura delosa</i>	2.7			X		0.9		2.5
			<i>Ostrocerca</i> prob. <i>truncata</i>			X					
			<i>Soyedina</i> sp.			X	X				0.2
		Peltoperlidae	<i>Peltoperla arcuata</i>	0.1	2.3	1.5	2.9		X	4.4	0.6
		Perlidae	<i>Acroneuria carolinensis</i>	1.0	2.3	0.5	0.8			3.7	3.1
			<i>Paragnetina</i> sp.	X							
Perlodidae			<i>Clioperla clio</i>			0.5					
			<i>Diploperla robusta</i>	X						0.7	
			<i>Isoperla holochlora</i>	X	X		1.4				0.6
			<i>Malirekus hastatus</i>	0.3		X	1.2				0.2
			<i>Yugus</i> sp.	1.3		0.5	X				



[illegible]



Class	Order	Family	Genus/Species	BEH				PBH			
				Sp	Su	F	W	Sp	Su	F	W
CRUSTACEA	Chironomidae		<i>Brilla</i> sp.		0.7					1.5	1.5
		<i>Constempellina</i> sp.				0.4					
		<i>Conchapelopia</i> sp.	1.0	0.7	1.0	0.4	1.7	6.1			
		<i>Corynoneura</i> sp.			0.5	0.4	0.3			X	1.0
		<i>Cricotopus trifascia</i> gp.	0.1								0.2
		<i>Dianesa</i> sp.				0.2				X	0.6
		<i>Diplocladius</i> sp.	0.3	0.3		0.6					1.0
		<i>Eukiefferiella</i> spp.	X								
		<i>Hydrobaenus</i> sp.									
		<i>Linnophyes</i> sp.			0.5						
		<i>Micropsectra</i> sp.			3.4	4.1					
		<i>Microtendipes pedellus</i> gp.	0.1				1.0			1.5	2.1
		<i>Microtendipes rydalsensis</i> gp.	0.1		X	X				1.5	0.2
		<i>Nilotanyus</i> sp.	0.4				0.3				
		<i>Orthocladius annectens</i>			0.5						
		<i>Parachaetocladius</i> sp.				0.2				0.7	0.6
		<i>Parametriochnemus lundbecki</i>	2.5	0.3	5.3	3.3	0.7			0.7	3.6
		<i>Polypedilum aviceps</i> gp.	X								
		<i>Polypedilum convictum</i> gp.	2.0	2.0	1.5			0.9			0.2
		<i>Polypedilum fallax</i> gp.									
	<i>Polypedilum haltarare</i> gp.			0.5							
	<i>Pothastia</i> sp.	0.1									
	<i>Rheotanytarsus</i> sp.	0.3	0.3								
	<i>Synorthocladius</i> sp.										
	<i>Tanytarsus</i> sp.	2.7	0.3	X		6.9			2.2	0.2	
	<i>Thienimanniella</i> sp.	0.3		1.5	X				X	0.4	
	<i>Tvetenia bavarica</i> gp.	0.3							1.5	1.0	
	<i>Tvetenia discoloripes</i> gp.								1.5	0.8	
	<i>Hemerodromia</i> sp.		0.3						0.7	0.8	
	<i>Dixa</i> sp.		0.3						0.7	0.2	
	<i>Prosimulium</i> sp.				0.8						
	<i>Simulium</i> sp.	0.3	0.3								
	<i>Hexatoma</i> sp.	2.5	5.6	2.9	1.9	2.4	0.9				
	<i>Dicranota</i> sp.		0.3	0.5	0.2						
	<i>Linnophila</i> sp.			1.0	0.2						
	<i>Pilaria</i> sp.	0.6									
	<i>Pseudolimmophila</i> sp.			2.9						0.4	
<i>Tipula</i> sp.	1.2	1.0	3.9			X		1.5	0.4		
Cambarid (immature)	0.3	0.7	X	0.4	0.3	1.8			0.2		
<i>Caecidotea</i> sp.	X										
Decapoda Isopoda											
Taxon Richness			66	42	56	62	33	44	54		
EPT Richness			35	20	29	35	19	23	26		



# Constructed Wetlands for Domestic Wastewater Treatment: Survey and Performance in Kentucky

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## ABSTRACT

Many residents of Kentucky live in rural areas not served by sanitary sewers. Wastewater must be disposed of in a manner that controls pathogens and other pollutants to protect the public health and minimize impacts on surface and groundwater. A promising solution to the wastewater problem in rural areas is an onsite subsurface flow (SSF) constructed wetland (CW) system for proper wastewater disposal. This paper summarizes results of an extensive CW survey of data collected from 67 county health department offices in Kentucky and the results of monitoring five existing SSF CW systems selected from Marshall, Letcher, and Fayette counties. Counties were grouped into those with <5 CW systems, 5 to 10, 10 to 50, 50 to 100, and >200; counties not using CW systems; and counties not having an exact wetland inventory or updated information. The most common plants used in SSF systems were cattails (*Typha latifolia*), reed canary grass (*Phalaris arundinacea*), blue water iris (*Iris versicolor*), softstem bulrush (*Scirpus validus*), pickerel weed (*Pontederia cordata*), and sweet flag (*Acorus calamus*). The selected SSF CW systems were monitored monthly for temperature, pH, dissolved oxygen (DO), biochemical oxygen demand (BOD<sub>5</sub>), total suspended solids (TSS), nitrate nitrogen (NO<sub>3</sub>-N), ammonia nitrogen (NH<sub>3</sub>-N), orthophosphate (PO<sub>4</sub> ion), and fecal coliform (FC) bacteria. Effluent BOD<sub>5</sub> and TSS concentrations averaged 12.3 and 19.8 mg/liter, indicating 68.6 and 84.1% removal, respectively. Ammonia nitrogen removal was low (51%), while nitrate levels dropped from 7.3 in influent wastewater to 1.8 mg/liter in effluent wastewater indicating 75% removal. Removal of orthophosphate ions (soluble phosphorus) averaged 46%. FC reduction averaged 94.7% but still far exceeded the reference level established by EPA. Dissolved oxygen increased from 0.53 mg/liter in influent wastewater to 1.35 mg/liter in the effluent discharge. The limited capacity for NH<sub>3</sub>-N, PO<sub>4</sub>, and FC removal necessitates further research into system design to increase efficiency.

## INTRODUCTION

In areas where municipal sewage treatment is not an option because the bedrock is near the surface and infiltration is impossible or where the water table is high and contaminants could be discharged directly to groundwater, onsite constructed wetland (CW) systems are appropriate. A CW system is defined as a designed and man-made complex that simulates natural wetlands for human use benefits. The character of the wetlands can be designed to fit the need presented by a particular wastewater to meet specific performance standards. Hammer (1989) indicated that each wetland should consist of five adjustable components: (1) substrates with various rates of hydraulic conductivity, (2) plants adapted to water-saturated anaerobic substrates, (3) water

column flowing in or above substrate's surface, (4) invertebrates and vertebrates, and (5) aerobic and anaerobic microbial populations. CW systems are becoming increasingly important as a technology for improving water quality. In SSF CW systems, wastewater is supposed to remain below the media surface; odor and insect vector problems are thus eliminated. In addition, no public-access problems exist because wastewater is not exposed.

The emergent aquatic plants used in SSF CW systems have the ability to transmit oxygen and other gases from the atmosphere through their leaves and stems above water to their root system, thereby producing an aerobic rhizosphere and thus increasing biological activity. In SSF systems, oxygen does not diffuse widely through the bed media but, at the



microsites adjacent to the root hairs, this oxygen is the major source in these systems. Further, its availability influences both the rate of BOD removal and nitrification of ammonia (Reed and Brown 1992). For optimum performance, therefore the roots must penetrate to the bottom of the media to increase availability of oxygen throughout the profile. The submerged portions of the plants can take up nutrients and other wastewater constituents and serve as the substrate for attached microbial growth. Morphology of aquatic plants varies widely, but for wastewater treatment purposes the plant type is determined by the depth of root and rhizome penetration.

Treatment of domestic sewage is a problem confronting small communities throughout the U.S. (Wolverton 1987a). SSF CW systems have the capacity to remove a large percentage of the total nitrogen and other pollutants in wastewater (Gersberg et al. 1983) and to satisfy regulatory effluent criteria established by the EPA (1993); they can be installed in a suitable location proximal to the home, taking advantage of land elevations. In all systems, the pollutants are removed by a complex variety of physical, chemical, and biological processes (Brix 1993). The major removal mechanism for nitrogen in CW systems is nitrification-denitrification (Gersberg et al. 1983). Oxygen plays an important role for many changes in wastewater composition. One oxygen source in these wetland beds is the leakage from roots of the aquatic plants. Therefore, it is essential to bring the wastewater into direct contact with the root zone. Brix and Schierup (1990) indicated that oxygen may also be supplied as a result of atmospheric air movement into the bed as the feedwater level falls during the flow-off period of an intermittent flow regime and also possibly as a result of flow around the gravel particles and through air-filled pores.

A major part of the treatment process for degradation of pollutants is attributed to the microorganisms living on and around root-systems of the plant in a symbiotic relationship (Wolverton 1987b). During microbial degradation of pollutants, metabolites are produced which the plants absorb and utilize along with nitrogen, phosphorus, and other minerals as a nutrient source while microorganisms use some or all metabolites released through plant roots as such a source. The synergistic effect

of this type of technology removes many of the substances contributing to BOD (e.g.,  $\text{NO}_3$ ,  $\text{NO}_2$ ,  $\text{NH}_4$ , and  $\text{PO}_4$  ions) from domestic sewage wastewaters. A  $\text{BOD}_5$  level of  $<20$  mg/liter and TSS level of  $<20$  mg/liter meet regulatory effluent criteria, according to EPA regulations (EPA 1993).

The increasing interest in CW systems for wastewater treatment in Kentucky is evidenced by the increasing number of wetlands being constructed. The number of operating systems has increased from fewer than five (Reed and Brown 1992) to over 4000 (Thom et al. 1998). The need for alternative wastewater treatment systems in Kentucky, due to the vulnerability of Kentucky's groundwater to pollution, had been indicated by many authors (Antonious and Byers 1996; Antonious et al. 1998a, 1998b; Steiner and Combs 1993). At least half of Kentucky's aquifer systems occur in karstic regions, which make these aquifers highly susceptible to contamination from the surface (Anonymous 1994).

The purpose of our paper is to highlight some of the important operational and monitoring issues that must be addressed by the scientific and engineering communities for improving constructed wetland performance in Kentucky. The main objectives of the study were (1) to establish a wetland inventory that contains type and number of aquatic plants used in CW systems in Kentucky, (2) to identify counties in Kentucky that employ CW technology, and (3) to provide information on the treatment efficiency of SSF CW systems used for single-home wastewater treatment.

## MATERIALS AND METHODS

### Wetland Survey

The survey data were obtained from different sources in Kentucky through personal collection, electronic files and printed reports, telephone contacts with the Cabinet of Human Resources, and the Department of Health Services/Environmental Management Branch in Frankfort. Each county health department was also contacted individually through telephone inquiries and was requested to submit all CW information within the respective district. According to the 1996 survey of Kentucky's 120 counties, 26 counties do not employ CWs. Within the remaining 94



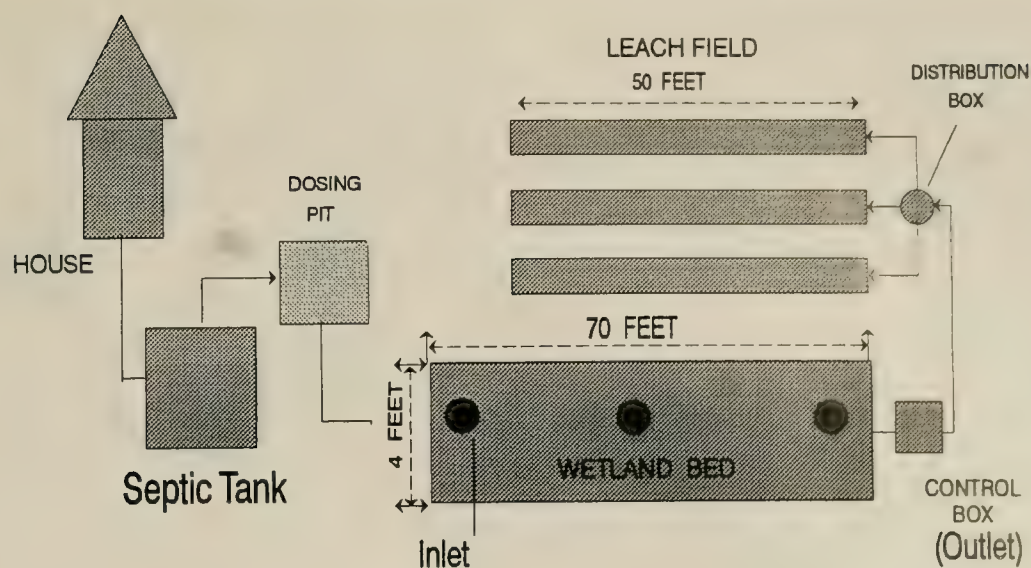


Figure 1. Schematic diagram of subsurface flow (SSF) constructed wetland systems in Kentucky for a three-bedroom house. Note that the dosing pit receives wastewater from the septic tank (500 gallon tank) and permits equal distribution throughout the system. Effluent from the wetland system enters a distribution piping of 50 × 3' long "leach field" before reaching the soil. Circular spots inside wetland beds denote sampling ports used for monitoring the system performance.

counties, 27 counties do not have an exact wetland inventory or updated information. A database (d-base III version 1.1) and a computer program were created to organize CW records in 67 counties. The information collected included the type, number, and distribution of aquatic plants used in the different counties and the number of CW systems per county.

### Wetland Design

The wetlands we monitored treat wastewater from single-family dwellings (each home has three bedrooms). The wetland cells were plastic-lined (21.34 m [70'] long, 1.22 m [4'] wide, and 0.46 m [18"] deep). This type of SSF, commonly called a rock-plant filter, was developed by National Aeronautic and Space Administration (NASA) at the National Space Technologies Laboratory in Mississippi (Wolverton 1987b). Generally, the trench was partially filled with No. 2 rock (crushed limestone) to a depth of 0.36 m to prevent clogging; the water level was maintained at 0.36 m; and the trench was then covered with No. 5 and 6 rock to a depth of 0.46 m. The inlet of the systems received wastewater from the septic tank. The estimated wastewater flow throughout each system was 1.36 m<sup>3</sup>/day (360 gallons/day). One plant (cattail, *Typha latifolia*) per 0.37 m<sup>2</sup> of bed surface was set out by

CW installers for optimum efficiency (Gersberg et al. 1983).

### Wetland Monitoring

Five CW systems were selected for monitoring from Marshall, Letcher, and Fayette counties. Influent and effluent wastewater from each wetland cell was sampled monthly from fixed sampling ports throughout the bed-rock system, the inlet (influent port), and from the discharge end (effluent port) of the wetland system (Figure 1). Samples were monitored for temperature, pH, and dissolved oxygen (DO) in the field and analyzed for biochemical oxygen demand (BOD<sub>5</sub>) in the 5-day test, total suspended solids (TSS), nitrate nitrogen (NO<sub>3</sub>-N), ammonia nitrogen (NH<sub>3</sub>-N), orthophosphate (PO<sub>4</sub> ion), and fecal coliform (FC) bacteria. The water quality parameters of the collected samples were analyzed at the Water Quality and Environmental Toxicology laboratory at Kentucky State University by the use of standard methods (APHA 1995). Ammonia (NH<sub>3</sub>-N) was determined by the selective ion electrode method 4500-F; BOD<sub>5</sub> by method 5210-B; nitrate (NO<sub>3</sub>-N) by method 4500-NO<sub>3</sub>-E; orthophosphate by method 4500-P-E; pH by method 4500-H; and total suspended solids by method 2540-D. FC bacterial analysis was conducted using the membrane filter standard method no. 9222 (APHA



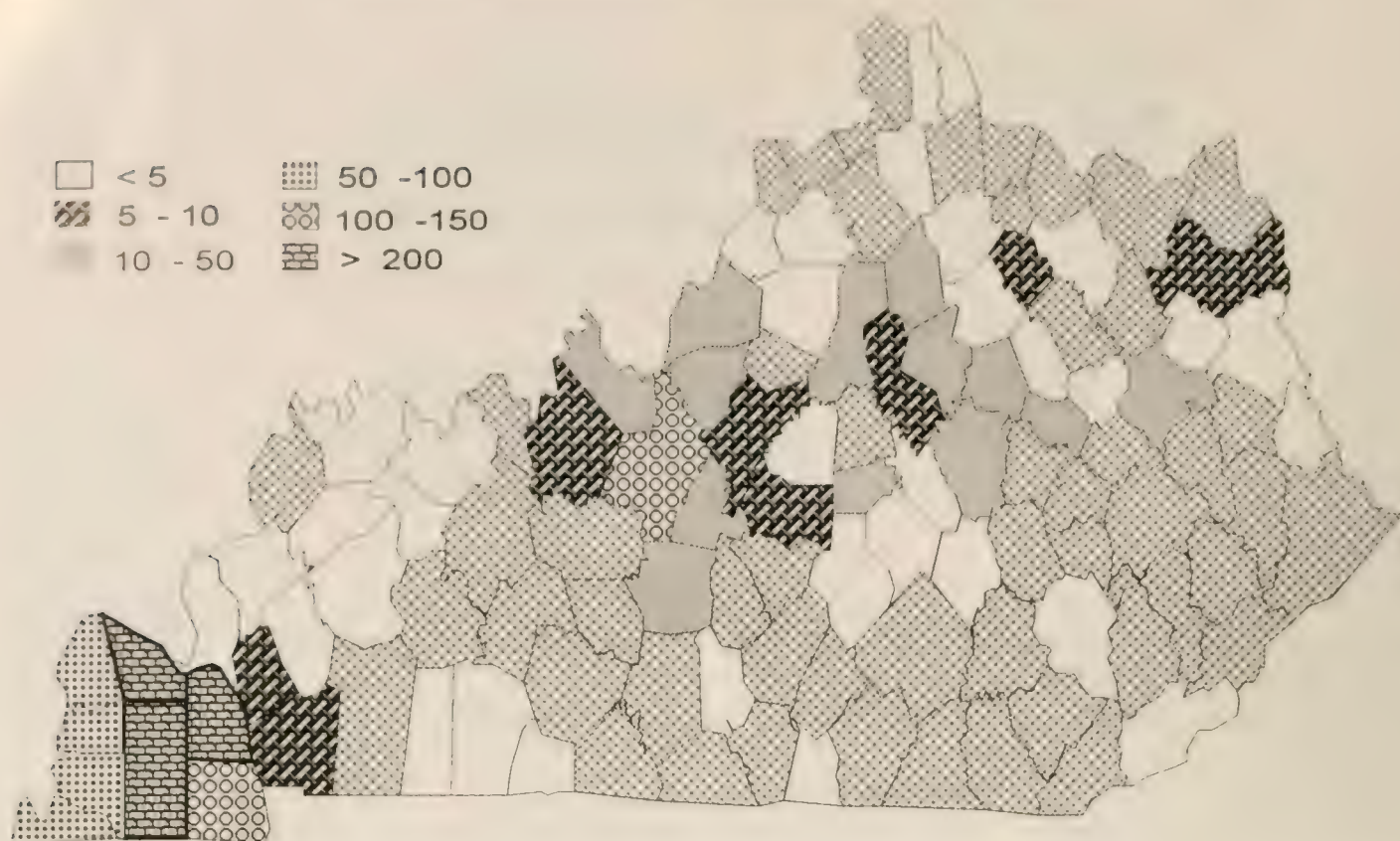


Figure 2. Number of subsurface flow (SSF) constructed wetland systems used for wastewater treatment from single-family dwellings and their distribution in Kentucky counties.

1995). All samples were collected from sampling ports 13 cm above the bottom of the system to avoid disturbing any sediment and were analyzed within 6 hours of sampling; accordingly, no preservatives were added to the samples. Data were analyzed for each water-quality parameter using analysis of variance (ANOVA) procedure (SAS Institute 1991). Means were compared using Duncan's LSD test (Snedecor and Cochran 1967).

## RESULTS AND DISCUSSION

Most of the SSF CW systems documented in Kentucky are in the southwestern counties (Figure 2). Because of the plant (hydrophyte) diversity present in a natural marsh, many types of plants have been utilized on CW systems. Table 1 shows the main species of plants used in SSF CW systems in Kentucky. The plants most commonly used are cattails, *Typha latifolia*; reed canary grass, *Phalaris arundinacea*; blue water iris, *Iris versicolor*; softstem bulrush, *Scirpus validus*; pickerel weed, *Pontederia cordata*; and sweet flag, *Acorus calamus*.

Overall BOD<sub>5</sub> average value dropped significantly ( $P < 0.05$ ) from 39.4 (influent port)

to 12.3 mg/liter at the discharge, an overall removal of 68.6%. TSS also decreased from 124.5 to 19.8 mg/liter at the discharge port, an 84% removal (Table 2). A common permit requirement of BOD<sub>5</sub> and TSS is 20 mg/liter reference level (EPA 1993).

Results also indicated that NO<sub>3</sub> concentration was significantly reduced from 7.3 mg/liter in influent port to 1.8 mg/liter at the discharge end of the system (75.2% removal). Because hydrogen ions are produced in the nitrification process, the pH of the system can drop. Below a pH of 6.5, the nitrification process is inhibited (Ogden 1994). Accordingly, to maintain the system performance sufficient alkalinity must be present or added to the system. For this reason, the limestone used as a bedrock in CW systems may aid in the nitrification process by adding alkalinity to the wastewater. However, data in Table 2 indicate that the pH of the influent and of the effluent wastewater was about 7 (neutral). Sutton (1990) reported that the alkalinity destruction rate is 7.1 mg alkalinity (as CaCO<sub>3</sub>) per mg of NH<sub>3</sub>-N (ammonia-nitrogen) oxidized.

The systems under study cannot satisfy the discharge requirements of FC removal. Over-



Table 1. Species of plants commonly used for subsurface flow (SSF) constructed wetland system for a three-bedroom house in Kentucky.

Plant	Scientific name	Number <sup>1</sup> of CW's	%
Cattails	<i>Typha latifolia</i>	136	24.9
Reed Canary Grass	<i>Phalaris arundinacea</i>	69	12.6
Blue Water Iris	<i>Iris versicolor</i>	67	12.2
Softstem Bulrush	<i>Scirpus validus</i>	64	11.7
Pickernelweed	<i>Pontederia cordata</i>	56	10.2
Sweet Flag	<i>Acorus calamus</i>	55	10.1
Tall Fescue	<i>Festuca elatior</i>	21	3.8
Arrow Head	<i>Sagittaria latifolia</i>	18	3.3
Yellow Water Iris	<i>Iris pseudacorus</i>	10	1.8
Calla Lily	<i>Zantedeschia aethiopica</i>	8	1.5
Cardinal Flower	<i>Lobelia cardinalis</i>	4	0.7
Buttercup	<i>Ranunculus flabellaris</i>	3	0.5
Other Plants <sup>2</sup>	—	36	6.6

<sup>1</sup> Number of SSF constructed wetland (CW) systems in Kentucky counties that correspond to each aquatic plant type (based on the 1996 survey conducted in this study). Total number of CW systems reported in the survey is 547.

<sup>2</sup> Other plants indicate plants used in CW systems other than those described in this table (based on the 1996 survey).

all average counts of FC were  $1.3 \times 10^6/100$  ml of wastewater in influent and  $6.8 \times 10^4/100$  ml at the discharge end of the system (effluent); however, these remaining levels (Table 2) are still far higher than the discharge requirements. Removal of FC bacteria using SSF CW systems is 97.8%. This, however, is not sufficient to meet the usual requirement of 200 colonies/100 ml of water (EPA 1993).

The ability of SSF CW systems to remove phosphorus (PO<sub>4</sub> ions) over the long term appears to be limited. The systems removed 46% of incoming phosphorus (Table 2). Phosphorus removal is dependent on substrate (bedrock) chemical composition. The limited capacity for phosphorus removal (Antonious and Byers 1996; Antonious et al. 1998b; Reed 1991) also could be due to the short retention time and limited interaction with the underlying substrate matrix.

Wetlands have the capacity to remove large percentages of total nitrogen in wastewater through various biological and chemical reactions. Chemoautotrophic nitrifying bacteria, mainly *Nitrobacter* and *Nitrosomonas*, oxidize ammonia (NH<sub>3</sub>) to nitrite (NO<sub>2</sub>) and nitrate (NO<sub>3</sub>), respectively. Nitrate and NO<sub>2</sub> are reduced by facultative bacteria to nitrous oxide (N<sub>2</sub>O) and nitrogen gas (N<sub>2</sub>) in the anaerobic denitrification process. Oxygen consumption in this process is due to the direct microbial oxidation of organic matter and oxidation of reduced substances (Davido and Conway 1991). Systems with good aeration will likely have most of the nitrogen in the nitrate form. Results of monitoring CW systems selected from Kentucky indicate that treatment of ammonia (NH<sub>3</sub>) is generally less successful than BOD and TSS, with only a few systems demonstrating reliable treatment (Antonious and

Table 2. Impact of a subsurface flow (SSF) constructed wetland system for a three-bedroom house on some wastewater quality parameters<sup>1</sup>.

Wastewater parameter	Influent	Effluent
TSS, mg/liter	124.52 ± 79.66 a	19.82 ± 10.10 b
BOD <sub>5</sub> , mg/liter	39.35 ± 19.27 a	12.34 ± 7.74 b
NH <sub>3</sub> -N, mg/liter	45.32 ± 24.40 a	22.20 ± 15.29 b
NO <sub>3</sub> -N, mg/liter	7.30 ± 2.06 a	1.81 ± 1.50 b
Fecal coliform (colonies/100 ml of water)	128465 ± 3466592 a	67687 ± 167919 a
Phosphate Ion (PO <sub>4</sub> ), mg/liter	3.2 ± 1.72 a	1.76 ± 1.04 b
Dissolved O <sub>2</sub> , mg/liter	0.53 ± 0.16 b	1.35 ± 0.69 a
pH	7.17 ± 0.23 a	7.08 ± 0.19 a
Temperature, °C	14.14 ± 1.26 a	13.00 ± 2.30 a

<sup>1</sup> Each value in the table is an average (n = 60) ± SE of 3 years sampling of 5 SSF constructed wetland systems monitored in Marshall, Letcher, and Fayette counties, Kentucky. Values within a row having different letters are significantly different from each other, using Duncan's LSD test (P < 0.05)



Byers 1996; Antonious et al. 1997). The constraint in treating  $\text{NH}_3$  may be due to inadequate oxygen in the wetland water column to support biological nitrification of  $\text{NH}_3$ . Most aquatic organisms need dissolved oxygen (DO) levels of 2 mg/liter or more to survive (Anonymous 1999). Choate et al. (1993) indicated also that inadequate DO for nitrification prevented the satisfactory treatment of  $\text{NH}_3$ . Levels of  $\text{NH}_3$  were significantly lower (22 mg/liter) in wastewater effluent than in influent (45 mg/liter) (Table 2). This decrease in  $\text{NH}_3$  level should be accompanied by an increase in  $\text{NO}_3$  level during the same sampling period if the nitrification process is occurring properly in the system. But what is clear from the percentage of  $\text{NH}_3$  removal (51%) is that the systems were not effective in reducing ammonia ( $\text{NH}_3$ ). In systems of good performance, the decrease in  $\text{NH}_3$  is due mainly to nitrification during aeration and the decrease in  $\text{NO}_3\text{-N}$  is due to denitrification and absorption by wetland plants.

If not treated, FC,  $\text{NH}_3$ , and  $\text{NO}_3$  move rapidly with wastewater through the soil into the groundwater, which may subsequently be used as a source of drinking water. In Kentucky, groundwater is an important source of rural domestic water (KDWP/DOW 1989). The Commonwealth is characterized by 50% karstic topography, which is particularly vulnerable to groundwater pollution. Surface activities impact this karstic groundwater through contamination by bacteria and nutrients such as FC,  $\text{PO}_4$ ,  $\text{NH}_3$ , and  $\text{NO}_3$  due to the presence of open conduits. The long-term effects of these contaminants can be very serious, especially if the water table is high or the soil layer thin. Tchobanoglous (1993) indicated that "The SSF systems that have become popular are essentially copied from European practice, with little or no attention to system hydraulics." As a consequence, a number of the CW treatment systems have failed to meet expectations. One of the major water-quality problems observed during our study is, with few exceptions, the inability of these systems to meet the discharge requirement limits for ammonia, which is believed to be due to the insufficient availability of oxygen to support the activity of the root-attached nitrifying bacteria. As a result, a large part of the bed becomes anaerobic, which precludes nitrification

of the wastewater flow passing through the anaerobic zone. Further research in the system design is needed to increase efficiency for FC,  $\text{NH}_3$ ,  $\text{NO}_3$ , and  $\text{PO}_4$  removal. Every failure of an onsite system and every improperly installed onsite system or improper disposal of wastewater can create a potential severe threat to the public and the environment. Wetland problems originate from poor design, installation, or homeowner maintenance. Installation and homeowner maintenance errors can be reduced through hands-on-training and education.

Our future objective at Kentucky State University/Water Quality Research is to combine different types of cells to achieve the desired performance (Antonious 1999). A peat/gravel filter, which has the important advantage of removing N and P from wastewater (Brooks and McKee 1992), can be used for nitrification. In addition, plant species, plant intensity, water depth, and rock type and porosity can be selected for optimum efficiency.

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## Nannyberry (*Viburnum lentago* L.; Caprifoliaceae) Excluded from the Kentucky Flora

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### ABSTRACT

According to historic and current botanical literature *Viburnum lentago* L., nannyberry, is an element of the Kentucky flora. A request for all herbarium specimens of *Viburnum* taxa collected in Kentucky was sent to 28 herbaria across eastern United States; 1023 specimens were received in response. Of the 20 labeled as *V. lentago*, none was referable to that species. Extensive fieldwork across all physiographic regions of Kentucky was undertaken to try to locate occurrences of *V. lentago*; no populations were found. *Viburnum lentago* is rejected as an element in the Kentucky flora. Diagnostic characteristics are provided to differentiate *V. lentago* from closely allied species with which it is often confused.

### INTRODUCTION

*Viburnum lentago* L., nannyberry, is a small tree or a large shrub distributed in north central and northeastern United States and adjacent Canada (Little 1976). Its habitat has been described as rocky hillsides, woodland edges, sedge meadows, stream banks, shrubby swamps, and swampy woods (Elias 1987; Voss 1996). Botanical literature from the early 1800s to the 1990s includes nannyberry as a component of the Kentucky flora (Browne and Athey 1992; Duncan and Duncan 1988; Garman 1913; Heineke 1987; KSNPC 1996; Linney 1882; McAtee 1956; M'Murtrie 1819; Short et al. 1833; Torrey and Gray 1841; Wharton and Barbour 1973). Several county or regional floras and generic treatments have also reported *V. lentago* as occurring in Kentucky (Greenwell 1935; Huffaker 1975; Kearney 1893; Rader 1976) and have cited herbarium specimens to document its occurrence. Conversely, *V. lentago* has been omitted or rejected for Kentucky during much of the same time period by other studies (Braun 1943; Elias 1987; Gunn 1968; Jones 1983; KSNPC 1997; Little 1976; Medley 1993). Thus the literature concerning presence or absence of *V. lentago* in the flora of Kentucky is ambiguous.

Early works by Short et al. (1833) and Torrey and Gray (1841) stated that *V. lentago* occurs in Kentucky. In the late 1800s Kearney (1893) also reported *V. lentago* as occurring in Kentucky and referred to vouchers of *V. lentago* collected in Harlan County. Additionally, Linney (1882) reported *V. lentago* in his survey results from five central and west central

Kentucky counties. In the early part of the 20th century Garman (1913) included *V. lentago* in his Kentucky woody plant list and ascribed it to Bath, Bell, and Fayette counties. Unfortunately, very few Garman collections survived the fire at the University of Kentucky in 1948 (Jones and Meadows 1948). In her flora of Nelson County, Greenwell (1935) also reported collecting *V. lentago* from a "limestone hillside, Cox Creek." In a statewide treatment, Braun (1943) reported nine species in the genus but omitted *V. lentago*. However, *V. lentago* was again included in the Kentucky flora by McAtee (1956). He reported the range of *V. lentago* as including the northeastern states and adjacent Canada with occasional populations in Ohio, Indiana, Illinois, and Kentucky (Ballard County). In the mid 1960s Gunn (1968) found no *V. lentago* plants in his survey of Jefferson, Bullitt, Hardin, Jefferson, Meade, Nelson, Oldham, Shelby, and Spencer counties. Gunn also rejected the claims of Greenwell and M'Murtrie because "no specimen was located to support the author's claim." Wharton and Barbour (1973) included *V. lentago* and noted that nannyberry is "often planted where a large shrub is desired, but in the wild it is rare in Kentucky." In a flora of a portion of Tygarts Creek in Carter County, Huffaker (1975) noted that *V. lentago* was "frequent." Rader (1976) conducting a biosystematic study of *V. rufidulum* Raf. and *V. prunifolium* L., reported collecting *V. lentago* in Franklin County, Kentucky. She also provided a range map showing *V. lentago* distributed across much of north central and eastern



Kentucky. In a nationwide treatment, Little (1976) mapped *V. lentago* as absent from Kentucky as did Jones (1983), who mapped *V. lentago* to the north and east of Kentucky at more northerly latitudes or, for populations east of Kentucky, at higher elevations. In the late 1980s, *V. lentago* was again mapped in Kentucky by Duncan and Duncan (1988). In the 1990s, *V. lentago* was also recorded as occurring in the Cumberland Plateau of Kentucky by Browne and Athey (1992). And as recently as 1996, nannyberry has been state listed as rare based on historical records (KSNPC 1996).

The goal of my study was to address the ambiguity in the literature regarding the occurrence of *V. lentago* in Kentucky. I reviewed all literature ascribing *V. lentago* to Kentucky, examined and annotated all obtainable herbarium vouchers collected in Kentucky and identified as *V. lentago*, and searched for populations of this species in likely habitats across the state.

#### MATERIALS AND METHODS

Literature reviews of botanical research in Kentucky were compiled by Davies (1953), Fuller (1979), Fuller et al. (1989), and Medley (1993). Sources within these compilations include primary research articles and floristic lists for the state that contain reference to *Viburnum* taxa, including *V. lentago*. A review of these sources was conducted to find references about *V. lentago* in previously published works. A review of the literature also revealed likely locations of specimens and was useful in directing loan requests.

Requests for loans of all *Viburnum* taxa collected in Kentucky were sent to 28 herbaria across eastern United States. I hoped to locate all *V. lentago* material as well as any *Viburnum* material mis-identified but referable to *V. lentago*. Herbaria nearby, or without lending policies, were visited in person. Habitat requirements of nannyberry were compiled from field guides and manuals (Deam 1932; Elias 1987; Gleason and Cronquist 1991; Voss 1996) to aid in my field search for *Viburnum* conducted from 1994 to 1998 across all physiographic regions of the state.

#### RESULTS AND DISCUSSION

Fifteen literature citations, including regional and county floras as well as state-wide plant

lists, were found to include *V. lentago* as part of the Kentucky flora. A lesser number of sources (seven), either did not mention or specifically rejected *V. lentago* from Kentucky. Only four of the sources cited collections, collection number, and herbarium of deposit of specimens labelled as *V. lentago* collected in Kentucky. Although most published sources indicated that *V. lentago* occurs in Kentucky, the number of these claims based on specimens were few. No previous reference was located that reviewed all specimens and literature sources to address the presence/absence ambiguity of nannyberry.

Loan requests and herbarium visits resulted in locating 1023 *Viburnum* specimens collected in Kentucky. Twenty vouchers labelled as *V. lentago* were received from 14 herbaria. Three purported *V. lentago* collections, cited in the literature and expected on loan, were not received. The *V. lentago* vouchers, collected in Kentucky between 1831 and 1988, represented material from the Bluegrass, Mississippian Plateau, Cumberland Plateau, and Coastal Plain physiographic regions of the state. All of these were referable to other species in *Viburnum* (Table 1), most commonly *V. prunifolium*.

Numerous problems were encountered when trying to assemble nannyberry vouchers for review. Many early Kentucky collections have been lost or exist only outside the state. In several cases, collections could not be located in herbaria. For example, although numbered specimens were cited in their publications, neither Rader's collection of *V. lentago* from Franklin County nor Greenwell's collection of *V. lentago* from Nelson County could be located. Additionally, Athey's collection of *V. lentago* from the Cumberland Plateau (cited in Browne and Athey 1992) could not be located at MEM or MUR. Additional efforts were taken to travel to the field sites specified by Greenwell and Rader and to relocate the *Viburnum* populations they may have sampled. The Nelson County search yielded collections of *V. prunifolium* (23 Apr 1995, Weckman and Weckman 1329 EKY) and *V. rufidulum* (23 Apr 1995, Weckman and Weckman 1328 EKY). The Franklin County search also produced collections of *V. rufidulum* (27 Sep 1998, Weckman and Weckman 4420 EKY) and *V. prunifolium* (27 Sep 1998, Weckman and



Table 1    Summary of Kentucky specimens misidentified as *Viburnum lentago* in various U.S. herbaria.

Collector and number	Year collected	Herbarium	Referable to:
H. H. Eaton 110	1831	PH	<i>Viburnum rufidulum</i>
J. S. Terrill s.n.	1892	KES	<i>V. prunifolium</i>
J. S. Terrill s.n.	1892	UK	<i>V. prunifolium</i>
H. Garman s.n.	1893	UK	<i>V. prunifolium</i>
T. H. Kearney 325	1893	A	<i>V. prunifolium</i>
T. H. Kearney 325	1893	GH	<i>V. prunifolium</i>
T. H. Kearney 325	1893	MO	<i>V. prunifolium</i>
T. H. Kearney 325	1893	NY	<i>V. prunifolium</i>
T. H. Kearney 325	1893	US	<i>V. prunifolium</i>
H. Garman s.n.	1900	UK	<i>V. prunifolium</i>
S. F. Price s.n.	1902	SIU	<i>V. rufidulum</i>
E. J. Palmer 16574	1919	A	<i>V. prunifolium</i>
E. J. Palmer 16574	1919	MO	<i>V. prunifolium</i>
E. J. Palmer 16574	1919	PH	<i>V. prunifolium</i>
R. A. Greenwell s.n.	1933	NA	could not be located
J. Grossman 615	1965	BEREA	<i>V. prunifolium</i>
C. W. Conn s.n.	1974	UK	<i>V. rufidulum</i>
W. Meijer et al. 1189	1974	MDKY	<i>V. prunifolium</i>
W. Huffaker et al. 805	1974	MDKY	<i>V. prunifolium</i>
W. Huffaker et al. 1155	1974	MDKY	<i>V. prunifolium</i>
L. L. Rader 1086	1975	TENN	could not be located
H. Bryan 2008	1988	EKY	<i>V. prunifolium</i>
R. Athey	?	MEM	could not be located

*Weckman 4410* EKY). Habitats at both sites were unlike those described for nannyberry populations. Insufficient locality data were available to attempt relocation of the Athey Cumberland Plateau site. However, 102 of the 120 counties of Kentucky across all physiographic regions of the state were searched. These field efforts resulted in ca. 375 collections of *Viburnum* from 98 Kentucky counties, but no native or adventive *V. lentago* was located.

It should be noted that correctly determined *V. lentago* material was received in a loan from MDKY. These collections (*Carr*, 22 Jun 1936, MDKY accession #00720, 00721), although labeled as collected in Kentucky, are rejected as representing Kentucky material. Most likely, these vouchers represent collections made in New York and mislabeled in processing subsequent to the death of the collector (Cranfill 1980, p. 63; H. Setser, Morehead State University, pers. comm., 5 Dec 1996; A. Risk, Morehead State University, pers. comm., 14 Nov 1997).

*Viburnum lentago* is cultivated in the state according to Medley (1993) in Bernheim Forest, Bullitt County, and Cherokee Park, Jefferson County. It is also known from cultivation in Madison County (Berea College cam-

pus, 10 Sep 1993, *Abbott 6274* BERE; Berea College campus, 3 Jun 1997, *Weckman 3505* EKY); in Powell County at Natural Bridge State Park (Hemlock Lodge, 7 May 1994, *Weckman and Weckman 686* EKY; Hemlock Lodge, 6 Jul 1995, *Weckman and Weckman 1723* EKY); and on the campus of Northern Kentucky University (J. W. Thieret, Northern Kentucky University, pers. comm., 31 Dec 1999). Because *V. lentago* is used in cultivation in Kentucky, it has the potential to naturalize at some point.

*Viburnum lentago* is most often confused with *V. prunifolium* and to a lesser degree with *V. rufidulum*. It may be differentiated from *V. rufidulum* by leaf and pubescence characteristics. Leaves in *V. rufidulum* are thick, elliptic in outline, rounded to retuse at the tip, and rusty-red tomentose on petioles, midrib, and buds. The leaves of *V. lentago*, in contrast, are thin, generally oblong in outline, long acuminate at the tip, and covered below with golden brown, peltate scales. Differences distinguishing *V. prunifolium* from *V. lentago* include acute leaf tips and lack of peltate scales on the lower leaf surface in *V. prunifolium*. Additionally, the calyx is stipitate on the fruit of *V. prunifolium*, but sessile in *V. lentago*.

*Viburnum lentago* is thus rejected as an in-



digenous element of the flora and is known only as a cultivated plant in Kentucky at this time. Managers of forest lands, preserves, and natural areas in Kentucky should be cognizant of the non-native status of this species in the state.

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# Some Algae of Land Between The Lakes, Kentucky and Tennessee, I. Chlorophyta

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## ABSTRACT

This paper lists 124 species of chlorophyte algae collected at Land Between The Lakes, Kentucky and Tennessee, and previously unreported or infrequently reported as occurring in Kentucky. Included are first Kentucky records for species of the genera *Chlorogonium* and *Docidium*.

## INTRODUCTION

Land Between The Lakes (LBL) is a 170,000-acre national recreation area in western Kentucky and Tennessee. LBL is an inland peninsula resulting from the impoundment of the Cumberland and Tennessee rivers, giving rise to Lake Barkley on the eastern border and Kentucky Lake on the western border. Over a period of several years (1989–1999) I secured periodic samples of the phytoplankton and metaphyton from Duncan Lake (Lyon County, Kentucky) and Energy Lake and Hematite Lake (Trigg County, Kentucky) and analyzed them for the algal taxa present. Phytoplankton was collected with a plankton net (80  $\mu$ m), and metaphyton was collected by hand squeezing aquatic macrophytes present in the lakes.

## CHECKLIST OF CHLOROPHYTA

Nomenclature follows that of Prescott (1962); Prescott, Croasdale, and Vinyard (1972, 1975, 1977); Prescott, Croasdale, Vinyard, and Bicudo (1981); Prescott, Bicudo, and Vinyard (1982); Croasdale, Bicudo, and Prescott (1983); Komarek and Fott (1983); Kadlubowska (1984); and Mrozinska (1985). All previous reports of chlorophyte algae from Kentucky are presented with descriptions and pertinent literature citations in Dillard (1989a, 1989b, 1990, 1991a, 1991b, 1993). Checklist entries preceded by a double asterisk represent new generic reports for Kentucky; those preceded by a single asterisk represent new reports of infra-generic rank. Others represent records of algal taxa infrequently reported from Kentucky.

## VOLVOCALES

**\*\****Chlorogonium euchlorum* Ehrenberg. Duncan and Energy lakes. Pl. 1, Fig. 1.

## TETRASPORALES

*Gloeocystis planctonica* (West & West) Lemmermann. Duncan, Energy, and Hematite lakes. Pl. 1, Fig. 2.

## CHLOROCOCCALES

*Planktosphaeria gelatinosa* G.M. Smith. Duncan, Energy, and Hematite lakes. Pl. 1, Fig. 3.

*Tetraedron minimum* (Braun) Hansgrig. Duncan, Energy, and Hematite lakes. Pl. 1, Fig. 4.

*T. regulare* Kuetzing. Energy and Hematite lakes. Pl. 1, Fig. 5.

*Schroederia setigera* (Schroeder) Lemmermann. Duncan Lake. Pl. 1, Fig. 6.

*Sphaerocystis schroeteri* Chodat. Duncan, Energy, and Hematite lakes. Pl. 1, Fig. 7.

*Oocystis parva* West & West. Duncan and Hematite lakes. Pl. 1, Fig. 8.

**\****O. solitaria* Wittrock. Duncan Lake. Pl. 1, Fig. 9.

*Eremosphaera viridis* DeBary. Duncan and Hematite lakes. Pl. 1, Fig. 10.

**\****Nephrocystium obesum* West & West. Duncan, Energy, and Hematite lakes. Pl. 8, Fig. 12.

*Kirchneriella lunaris* (Kircher) Moebius. Duncan and Hematite lakes. Pl. 1, Fig. 11.

*K. obesa* (West) Schmidle. Duncan and Hematite lakes. Pl. 1, Fig. 12.

*Selenastrum gracile* Reinsch. Duncan and Hematite lakes. Pl. 1, Fig. 13.

*Ankistrodesmus falcatus* (Corda) Ralfs. Dun-



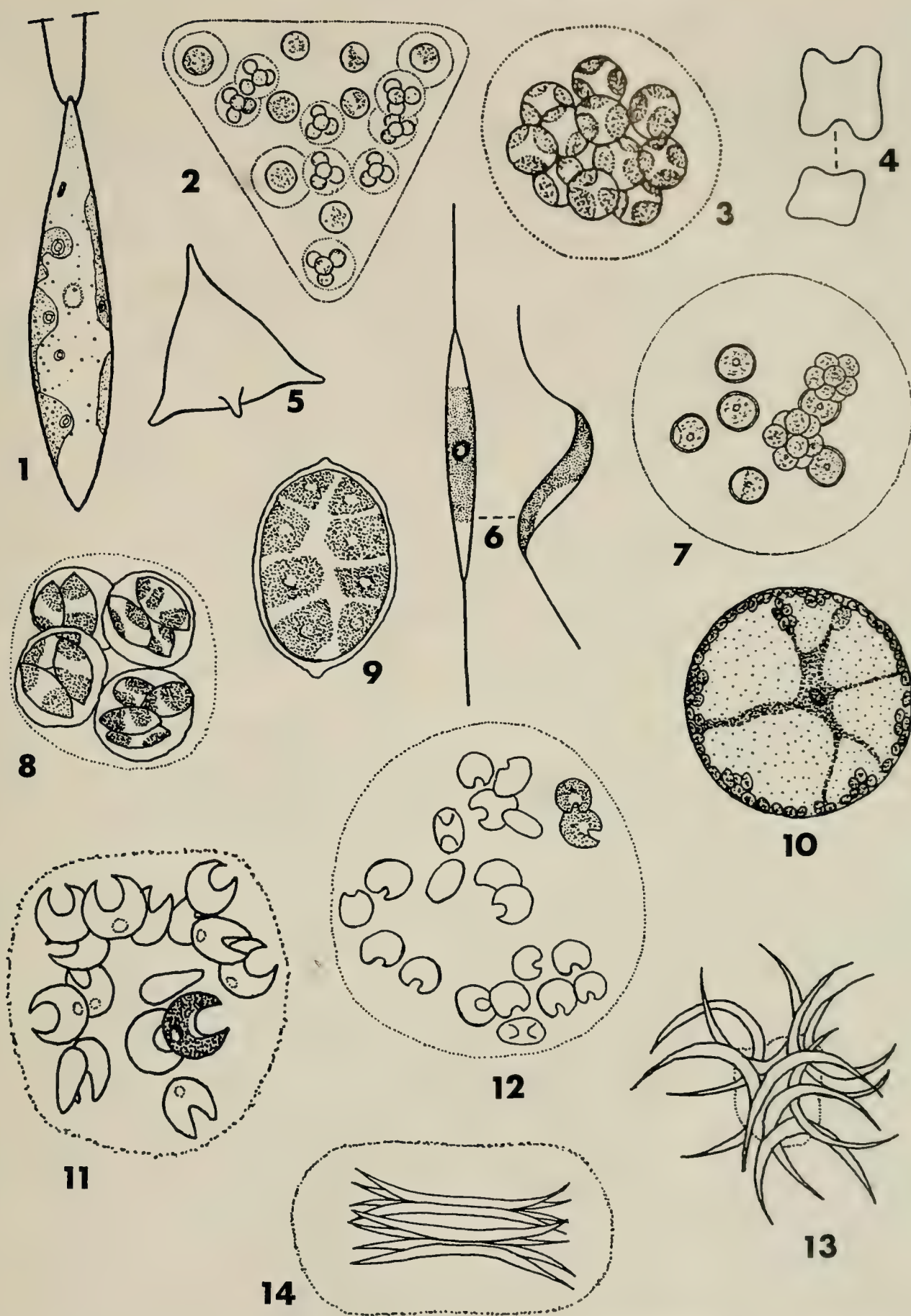


Plate 1. 1. *Chlorogonium euchlorum*; 2. *Gloeocystis planctonica*; 3. *Planktosphaeria gelatinosa*; 4. *Tetraedron minimum*; 5. *T. regulare*; 6. *Schroederia setigera*; 7. *Sphaerocystis Schroeteri*; 8. *Oocystis parva*; 9. *O. solitaria*; 10. *Eremosphaera viridis*; 11. *Kirchneriella lunaris*; 12. *K. obesa*; 13. *Selenastrum gracile*; 14. *Ankistrodesmus falcatus*.



can, Energy, and Hematite lakes. Pl. 1, Fig. 14.

*A. spiralis* (Turner) Lemmermann. Duncan and Energy lakes. Pl. 2, Fig. 1.

*Closteriopsis longissima* Lemmermann. Duncan and Hematite lakes. Pl. 2, Fig. 2.

*Quadrigula chodatii* (Tanner-Fullman) G.M. Smith. Duncan lake. Pl. 2, Fig. 3.

*Golenkinia radiata* (Chodat) Wille. Duncan and Hematite lakes. Pl. 2, Fig. 4.

*Botryococcus braunii* Kuetzing. Duncan, Energy, and Hematite lakes. Pl. 2, Fig. 5.

*Dictyosphaerium pulchellum* Wood. Duncan and Hematite lakes. Pl. 2, Fig. 6.

*Coelastrum cambricum* Archer. Hematite Lake. Pl. 2, Fig. 7.

*C. microporum* Naegeli. Energy and Hematite lakes. Pl. 2, Fig. 8.

*Crucigenia tetrapedia* (Kirchner) West & West. Duncan and Hematite lakes. Pl. 2, Fig. 9.

\**Scenedesmus bicaudatus* (Hansgirg) Chodat. Hematite Lake. Pl. 2, Fig. 10.

*S. dimorphus* (Turpin) Kuetzing. Duncan, Energy, and Hematite lakes. Pl. 2, Fig. 11.

*S. obtusus* Meyen. Duncan, Energy, and Hematite lakes. Pl. 2, Fig. 12.

*S. quadricauda* (Turpin) Brebisson. Duncan, Energy, and Hematite lakes. Pl. 2, Fig. 13.

*Actinastrum gracillimum* G.M. Smith. Hematite Lake. Pl. 2, Fig. 14.

\**Sorastrum americanum* (Bohlin) Schmidle. Duncan Lake. Pl. 2, Fig. 15.

*Pediastrum boryanum* (Turpin) Meneghini. Duncan, Energy, and Hematite lakes. Pl. 2, Fig. 16.

*P. duplex* Meyen. Duncan, Energy, and Hematite lakes. Pl. 2, Fig. 17.

*P. simplex* Meyen. Duncan and Energy lakes. Pl. 2, Fig. 18.

#### MICROSPORALES

\**Microspora pachyderma* (Wille) Lagerheim. Hematite Lake. Pl. 3, Fig. 1.

#### CHAETOPHORALES

*Coleochaete orbicularis* Pringsheim. Hematite Lake. Pl. 3, Fig. 2.

*C. scutata* Brebisson. Hematite Lake. Pl. 3, Fig. 3.

#### OEDOGONIALES

Several species of *Oedogonium* and *Bulbochaete* were collected from Duncan, Energy,

and Hematite lakes but could not be identified to the species level due to the absence of mature oospores.

\**Oedogonium boschii* (LeClerc) Wittrock. Duncan Lake. Pl. 3, Fig. 4.

\**O. capilliforme* Kuetzing. Duncan Lake. Pl. 3, Fig. 5.

\**O. cardiacum* (Hassall) Wittrock. Hematite Lake. Pl. 3, Fig. 6.

*O. grande* Kuetzing. Hematite Lake. Pl. 3, Fig. 7.

*Bulbochaete varians* Wittrock. Duncan Lake. Pl. 3, Fig. 8.

#### ZYGNEMATALES

Several species of *Spirogyra*, *Mougeotia*, and *Zygnema* were collected from Duncan, Energy, and Hematite lakes but could not be identified to the species level due to the absence of mature zygospores.

*Spirogyra communis* (Hassall) Kuetzing. Duncan and Hematite lakes. Pl. 3, Fig. 9.

*S. pratensis* Transeau. Energy Lake. Pl. 3, Fig. 10.

*S. varians* (Hassall) Kuetzing. Hematite Lake. Pl. 3, Fig. 11.

\**Mougeotia boodlei* (West & West) Collins. Hematite Lake. Pl. 3, Fig. 12.

*M. sphaerocarpa* Wolle. Energy Lake. Pl. 3, Fig. 13.

\**Zygnema decussatum* (Vaucher) Agardh. Duncan Lake. Pl. 3, Fig. 14.

*Spirotaenia condensata* Brebisson. Hematite Lake. Pl. 4, Fig. 1.

*Netrium digitus* (Ehrenberg) Itzigson & Rothe. Duncan and Hematite lakes. Pl. 4, Fig. 2.

\**Gonatozygon brebissonii* DeBary. Duncan, Energy, and Hematite lakes. Pl. 4, Fig. 3.

*Penium margaritaceum* (Ehrenberg) Brebisson. Duncan, Energy, and Hematite lakes. Pl. 4, Fig. 4.

*Closterium abruptum* West. Duncan Lake. Pl. 4, Fig. 5.

*C. ehrenbergii* Meneghini. Duncan and Hematite lakes. Pl. 4, Fig. 6.

*C. setaceum* Ehrenberg. Duncan and Hematite lakes. Pl. 4, Fig. 7.

\*\**Docidium baculum* Brebisson. Duncan and Hematite lakes. Pl. 4, Fig. 8.

\*\**D. undulatum* Bailey. Hematite Lake. Pl. 4, Fig. 9.



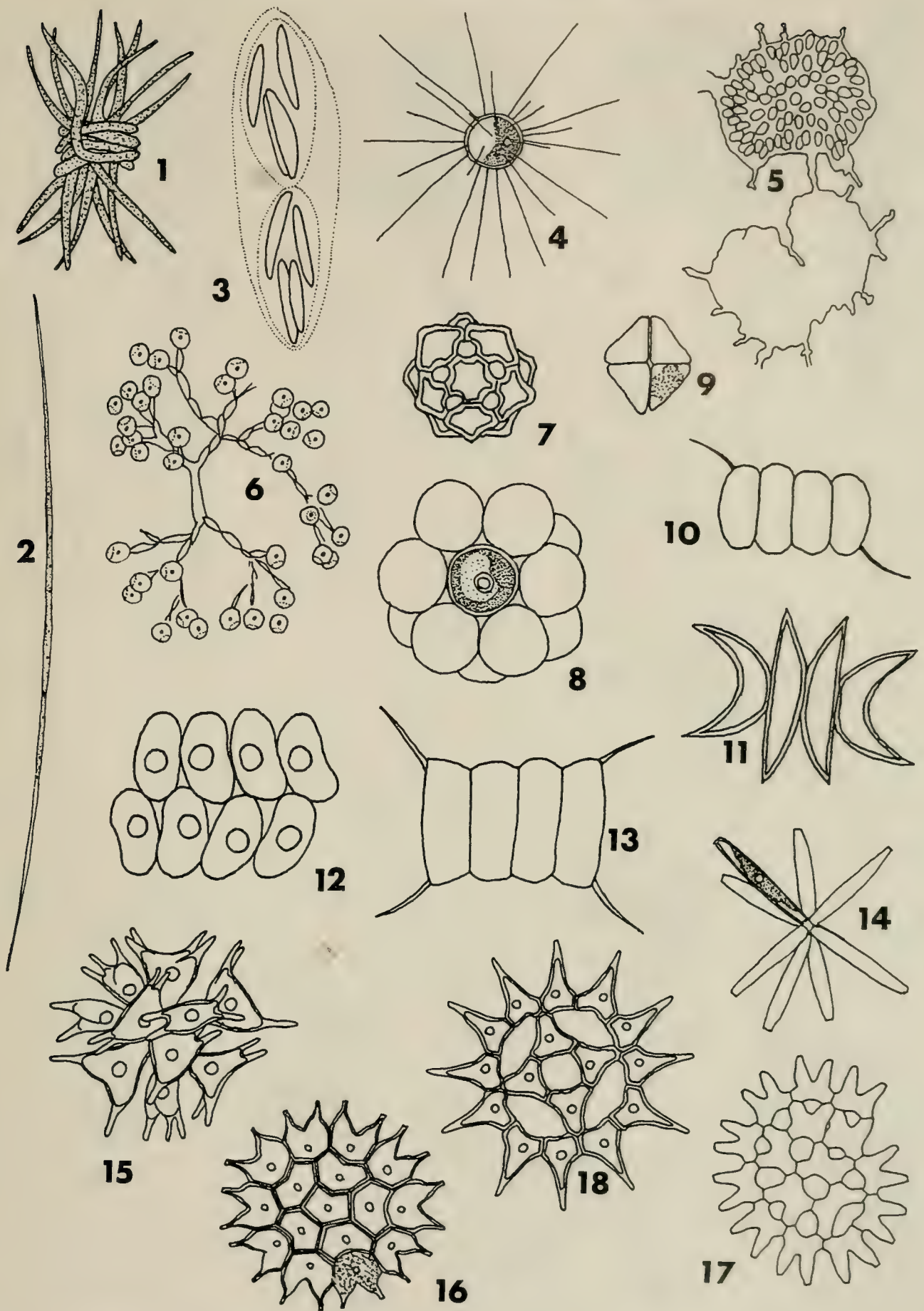


Plate 2. 1. *Ankistrodesmus spiralis*; 2. *Closteriopsis longissima*; 3. *Quadrigula chodatii*; 4. *Golenkinia radiata*; 5. *Botryococcus braunii*; 6. *Dictyosphaerium pulchellum*; 7. *Coelastrum cambricum*; 8. *C. microporum*; 9. *Crucigenia tetrapedia*; 10. *Scenedesmus bicaudatus*; 11. *S. dimorphus*; 12. *S. obtusus*; 13. *S. quadricauda*; 14. *Actinastrum gracillimum*; 15. *Sorastrum americanum*; 16. *Pediastrum boryanum*; 17. *P. duplex*; 18. *P. simplex*.



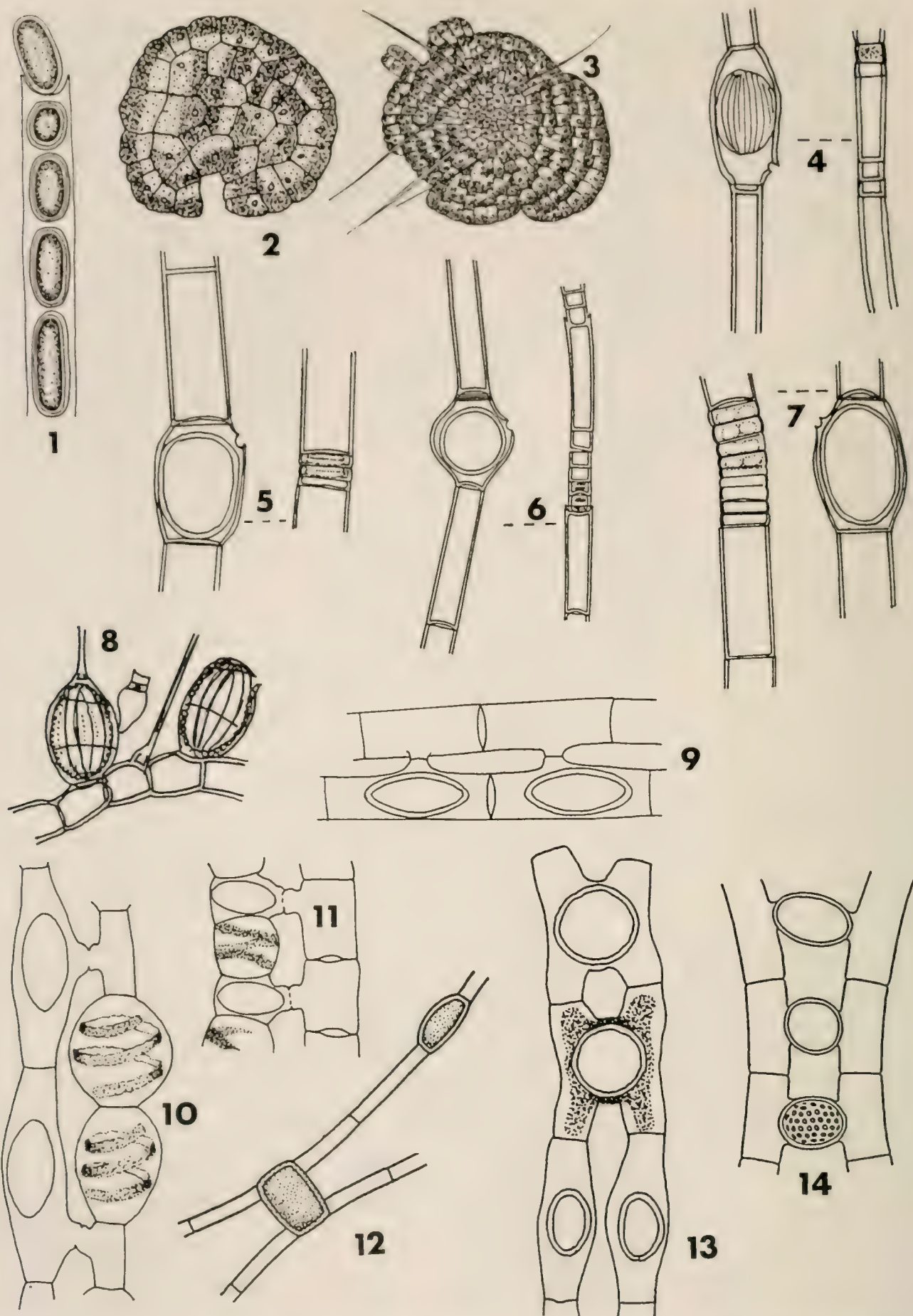


Plate 3. 1. *Microspora pachyderma*; 2. *Coleochaete orbicularis*; 3. *C. scutata*; 4. *Oedogonium boscii*; 5. *O. capilliforme*; 6. *O. cardiacum*; 7. *O. grande*; 8. *Bulbochaete varians*; 9. *Spirogyra communis*; 10. *S. pratensis*; 11. *S. varians*; 12. *Mougeotia hoodlei*; 13. *M. sphaerocarpa*; 14. *Zygnema decussatum*.



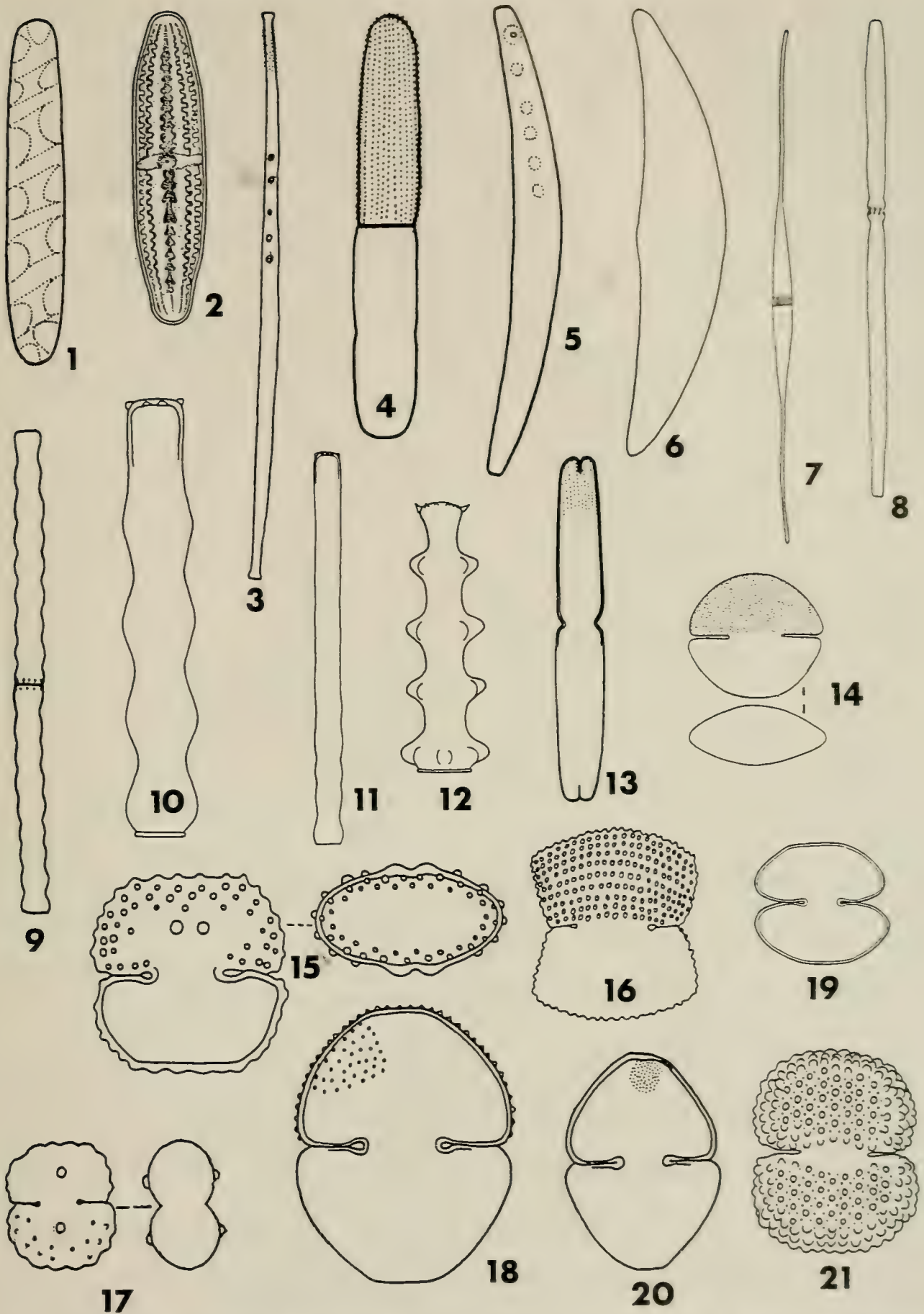


Plate 4. 1. *Spirotaenia condensata*; 2. *Netrium digitus*; 3. *Gonatozygon brebissonii*; 4. *Penium margaritaceum*; 5. *Closterium abruptum*; 6. *C. ehrenbergii*; 7. *C. setaceum*; 8. *Docidium baculum*; 9. *D. undulatum*; 10. *Pleurotaenium constrictum*; 11. *P. ehrenbergii*; 12. *P. nodosum*; 13. *Tetmemorus brebissonii*; 14. *Cosmarium baileyi*; 15. *C. bipunctatum*; 16. *C. biretum*; 17. *C. blyttii*; 18. *C. botrytis*; 19. *C. depressum*; 20. *C. granatum*; 21. *C. margaritatum*.



- \**Pleurotaenium constrictum* (Bailey) Wood. Hematite Lake. Pl. 4, Fig. 10.
- P. ehrenbergii* (Brebisson) DeBary. Duncan and Hematite lakes. Pl. 4, Fig. 11.
- P. nodosum* (Bailey) Lundell. Hematite Lake. Pl. 4, Fig. 12.
- Tetmemorus brebissonii* (Meneghini) Ralfs. Duncan Lake. Pl. 4, Fig. 13.
- Cosmarium baileyi* Wolle. Duncan and Energy lakes. Pl. 4, Fig. 14.
- \**C. bipunctatum* Boergesen. Hematite Lake. Pl. 4, Fig. 15.
- C. biretum* (Brebisson) Ralfs. Duncan Lake. Pl. 4, Fig. 16.
- C. blyttii* Wille. Duncan and Hematite lakes. Pl. 4, Fig. 17.
- C. botrytis* (Meneghini) Ralfs. Duncan Lake. Pl. 4, Fig. 18.
- C. depressum* (Naegeli) Lundell. Duncan, Energy, and Hematite lakes. Pl. 4, Fig. 19.
- C. granatum* Brebisson. Hematite Lake. Pl. 4, Fig. 20.
- C. margaritatum* (Lundell) Roy & Bissett. Duncan, Energy, and Hematite lakes. Pl. 4, Fig. 21.
- C. meneghinii* Brebisson. Duncan and Hematite lakes. Pl. 5, Fig. 1.
- C. moniliforme* (Turpin) Ralfs. Energy and Hematite lakes. Pl. 5, Fig. 2.
- \**C. nymannianum* Grunow. Duncan Lake. Pl. 5, Fig. 3.
- C. obtusatum* Schmidle. Duncan and Energy lakes. Pl. 5, Fig. 4.
- \**C. orthostichum* Lundell. Hematite Lake. Pl. 5, Fig. 5.
- C. ovale* Ralfs. Duncan, Energy, and Hematite lakes. Pl. 5, Fig. 6.
- \**C. phaseolus* Brebisson. Hematite Lake. Pl. 5, Fig. 7.
- \**C. porrectum* Nordstedt. Hematite Lake. Pl. 5, Fig. 8.
- C. portianum* Archer. Duncan and Hematite lakes. Pl. 5, Fig. 9.
- C. pyramidatum* Brebisson. Duncan and Hematite lakes. Pl. 5, Fig. 10.
- C. subtumidum* Nordstedt. Hematite Lake. Pl. 5, Fig. 11.
- C. turpinii* Brebisson. Duncan and Hematite lakes. Pl. 5, Fig. 12.
- Cosmocladium pusillum* Hilse. Duncan Lake. Pl. 5, Fig. 13.
- Arthrodesmus convergens* Ehrenberg. Duncan and Hematite lakes. Pl. 5, Fig. 14.
- A. extensus* (Borge) Hirano. Energy Lake. Pl. 5, Fig. 15.
- A. octocornis* Ehrenberg. Hematite Lake. Pl. 5, Fig. 16.
- Xanthidium antilopaeum* (Brebisson) Kuetzing. Duncan Lake. Pl. 6, Fig. 1.
- \**X. armatum* (Brebisson) Rabenhorst. Hematite Lake. Pl. 6, Fig. 2.
- Staurastrum alternans* (Brebisson) Ralfs. Duncan, Energy, and Hematite lakes. Pl. 6, Fig. 3.
- S. arctiscon* (Ehrenberg) Lundell. Duncan and Hematite lakes. Pl. 6, Fig. 4.
- S. botryophilum* Wolle. Duncan and Hematite lakes. Pl. 6, Fig. 5.
- \**S. brasiliense* Nordstedt. Duncan and Hematite lakes. Pl. 6, Fig. 6.
- S. chaetoceros* (Schroeder) G.M. Smith. Hematite Lake. Pl. 6, Fig. 7.
- \**S. crenulatum* (Naegeli) Delponte. Hematite Lake. Pl. 6, Fig. 8.
- \**S. curvatum* West. Duncan and Hematite lakes. Pl. 6, Fig. 9.
- \**S. dickiei* Ralfs. Duncan Lake. Pl. 6, Fig. 10.
- \**S. hexacerum* (Ehrenberg) Wittrock. Hematite Lake. Pl. 6, Fig. 11.
- S. leptocladum* Nordstedt. Hematite Lake. Pl. 6, Fig. 12.
- \**S. limneticum* Schmidle. Duncan and Energy lakes. Pl. 6, Fig. 13.
- S. setigerum* Cleve. Duncan and Hematite lakes. Pl. 6, Fig. 14.
- \**Euastrum abruptum* Nordstedt. Hematite Lake. Pl. 7, Fig. 1.
- \**E. affine* Ralfs. Hematite Lake. Pl. 7, Fig. 2.
- E. ansatum* Ehrenberg. Duncan Lake. Pl. 7, Fig. 3.
- E. binale* (Turpin) Ehrenberg. Duncan, Energy, and Hematite lakes. Pl. 7, Fig. 4.
- E. denticulatum* (Kirchner) Gay. Duncan and Energy lakes. Pl. 7, Fig. 5.
- E. didelta* (Turpin) Ralfs. Hematite Lake. Pl. 7, Fig. 6.
- E. elegans* (Brebisson) Ralfs. Duncan and Hematite lakes. Pl. 7, Fig. 7.
- \**E. evolutum* (Nordstedt) West & West. Duncan Lake. Pl. 7, Fig. 8.
- E. insulare* (Wittrock) Roy. Energy Lake. Pl. 7, Fig. 9.
- E. verrucosum* Ehrenberg. Duncan, Energy, and Hematite lakes. Pl. 7, Fig. 10.
- Micrasterias americana* (Ehrenberg) Ralfs. Duncan and Hematite lakes. Pl. 7, Fig. 11.



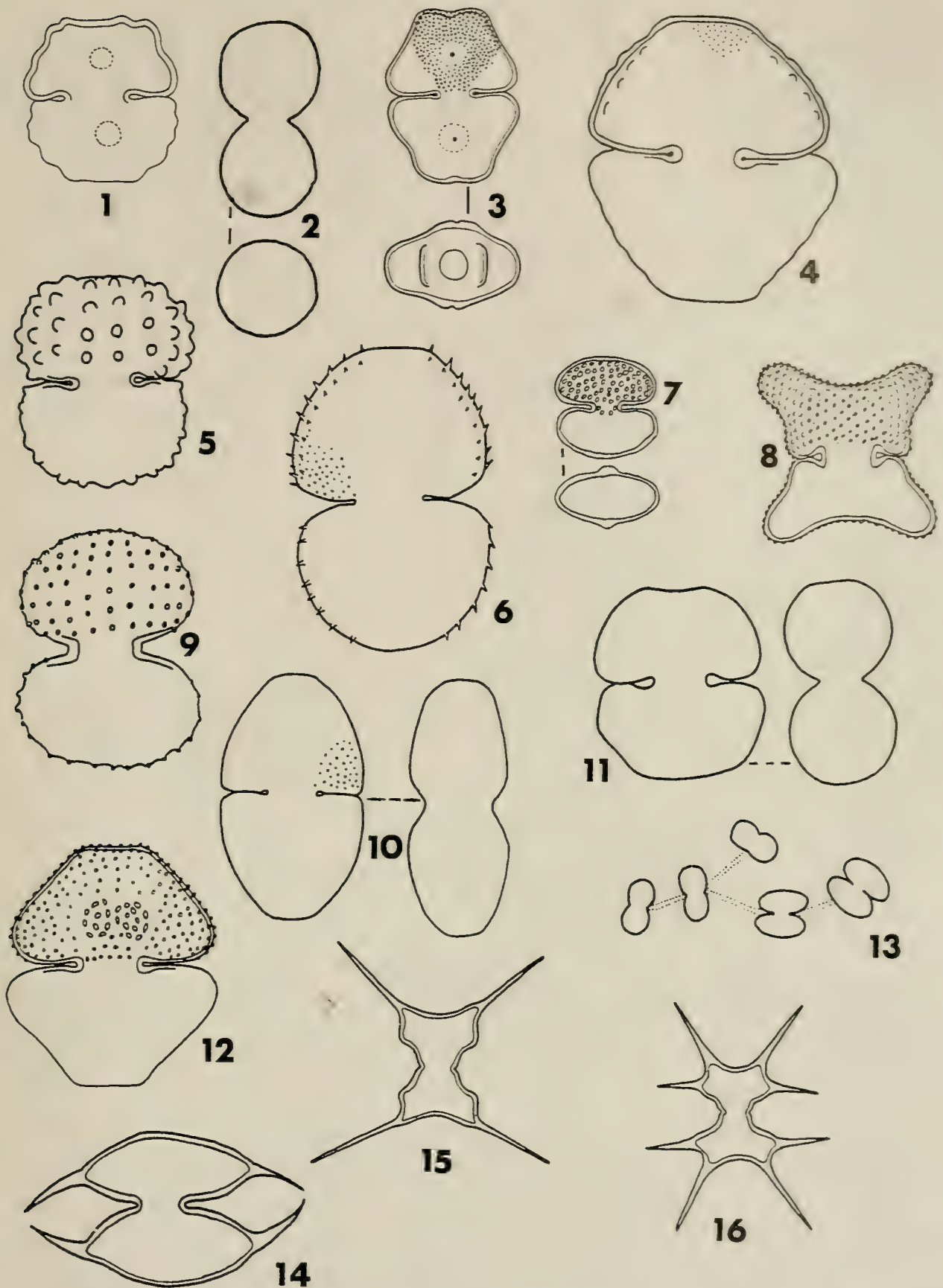


Plate 5. 1. *Cosmarium meneghinii*; 2. *C. moniliforme*; 3. *C. nymannianum*; 4. *C. obtusatum*; 5. *C. orthostichum*; 6. *C. ovale*; 7. *C. phaseolus*; 8. *C. porrectum*; 9. *C. portianum*; 10. *C. pyramidatum*; 11. *C. subtumidum*; 12. *C. turpinii*; 13. *Cosmocladium pusillum*; 14. *Arthrodesmus convergens*; 15. *A. extensus*; 16. *A. octocornis*.



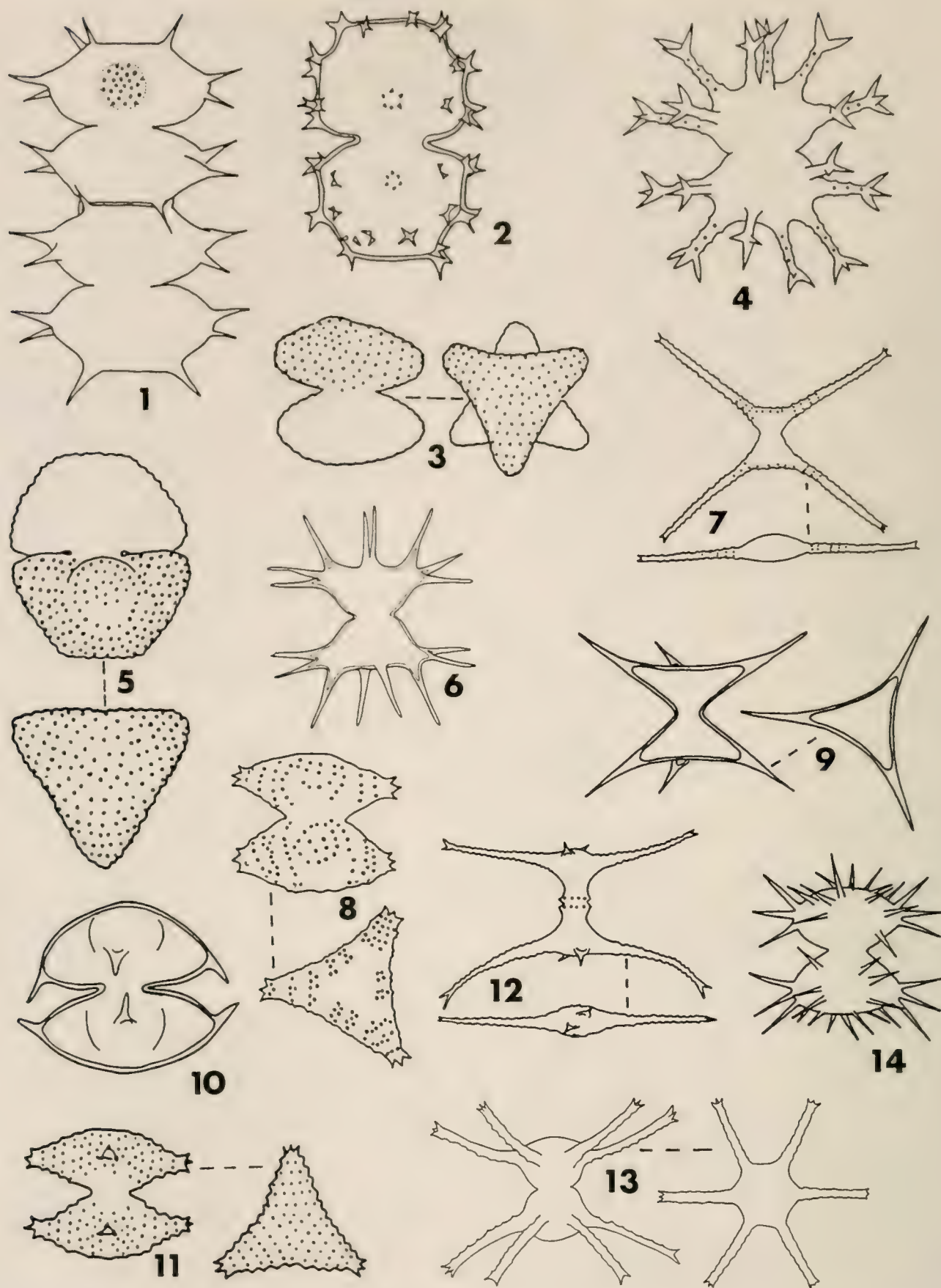


Plate 6. 1. *Xanthidium antilopaeum*; 2. *X. armatum*; 3. *Staurastrum alternans*; 4. *S. arcticon*; 5. *S. botryophilum*; 6. *S. brasiliense*; 7. *S. chaetoceros*; 8. *S. crenulatum*; 9. *S. curvatum*; 10. *S. dickiei*; 11. *S. hexacerum*; 12. *S. leptocladum*; 13. *S. limneticum*; 14. *S. setigerum*.



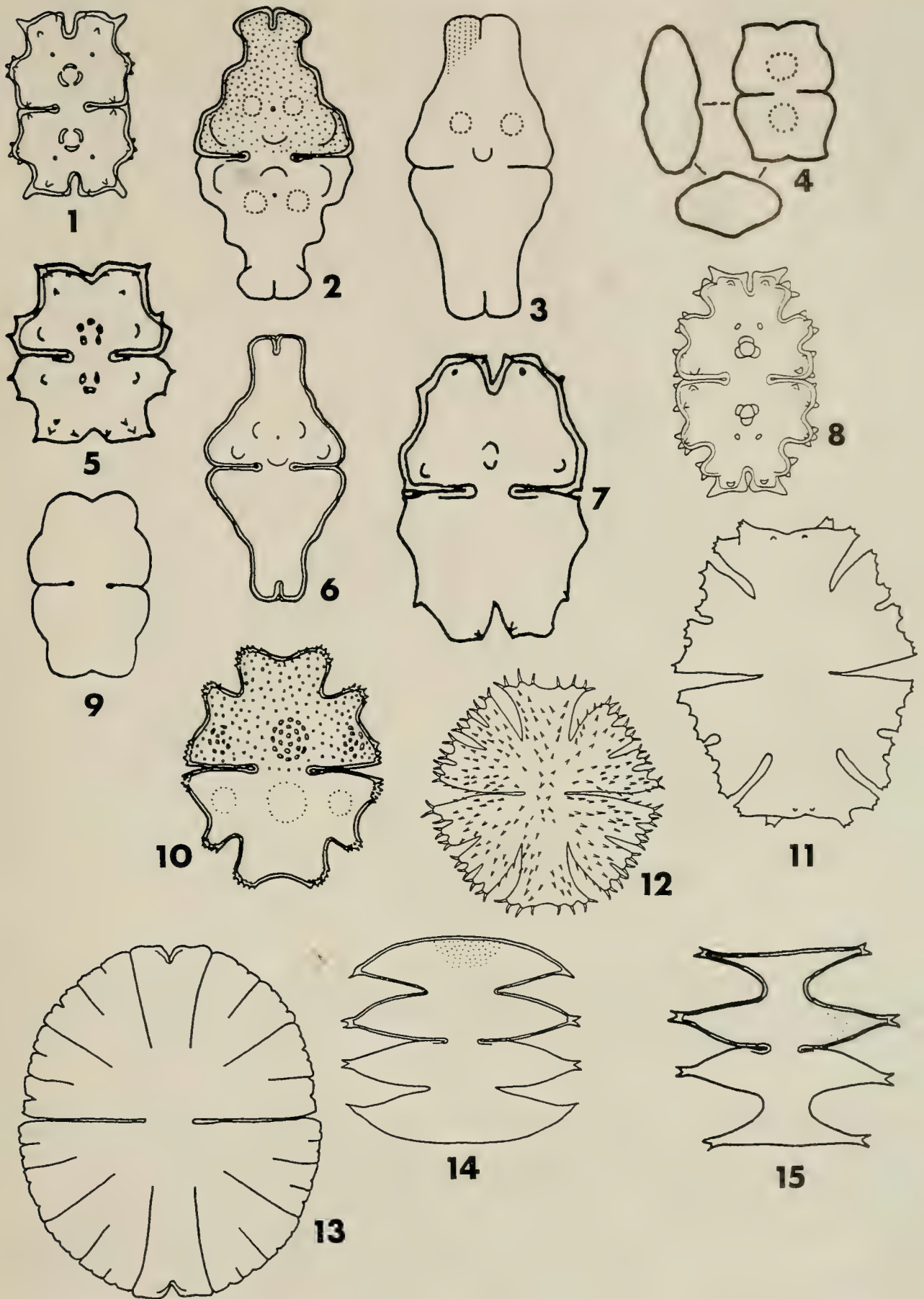


Plate 7. 1. *Euastrum abruptum*; 2. *E. affine*; 3. *E. ansatum*; 4. *E. binale*; 5. *E. denticulatum*; 6. *E. didelta*; 7. *E. elegans*; 8. *E. evolutum*; 9. *E. insulare*; 10. *E. verrucosum*; 11. *Micrasterias americana*; 12. *M. apiculata*; 13. *M. denticulata*; 14. *M. laticeps*; 15. *M. pinnatifida*.



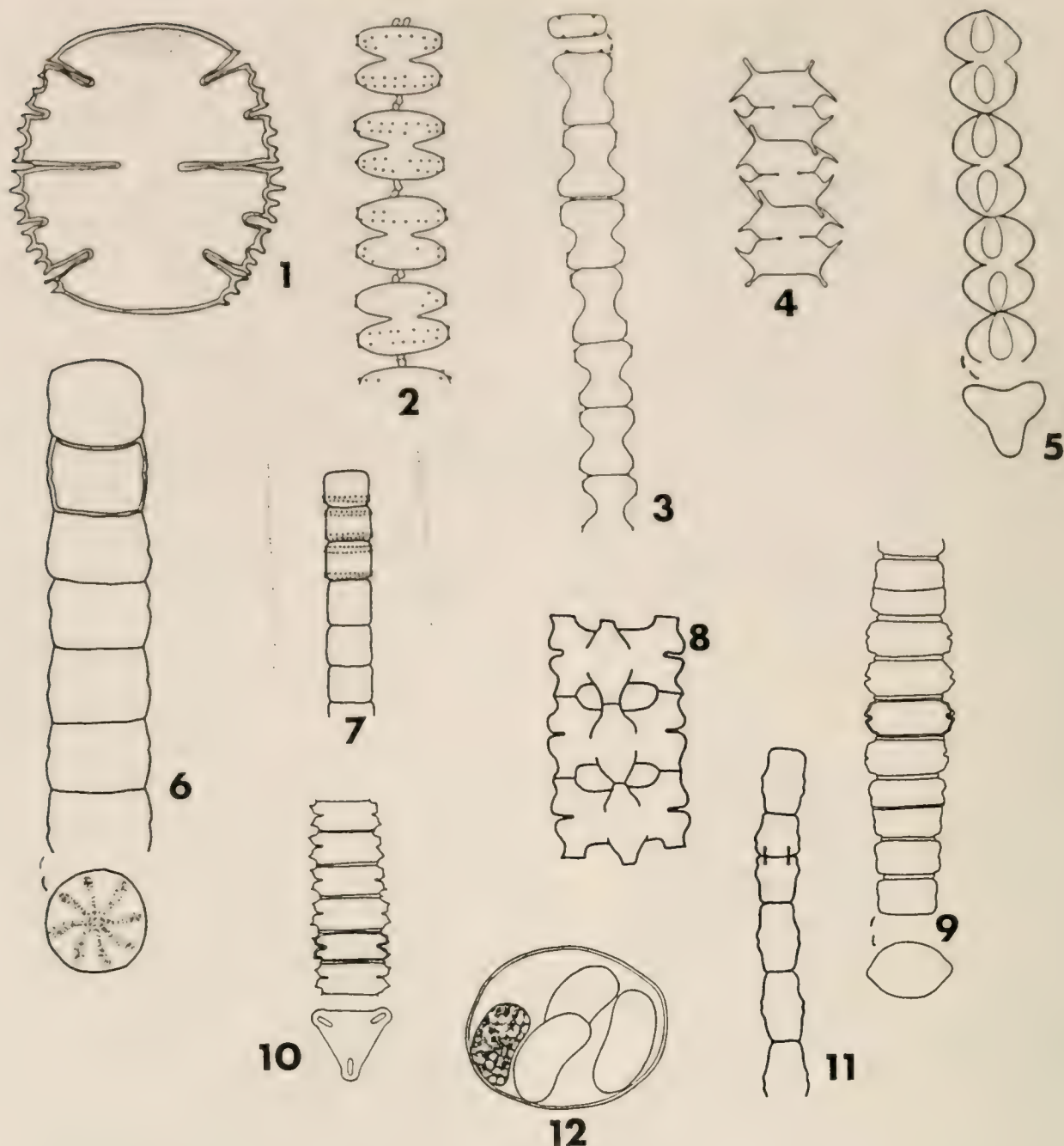


Plate 8. 1. *Micrasterias truncata*; 2. *Sphaerosozma aubertianum*; 3. *Teilingia excavata*; 4. *Onychonema laeve*; 5. *Spondylosium moniliforme*; 6. *Hyalotheca dissiliens*; 7. *H. mucosa*; 8. *Desmidium aptogonum*; 9. *D. grevillii*; 10. *D. swartzii*; 11. *Bambusina brebissonii*; 12. *Nephrocytium obesum*.

\**M. apiculata* (Ehrenberg) Ralfs. Duncan Lake. Pl. 7, Fig. 12.

*M. denticulata* Brebisson. Hematite and Energy lakes. Pl. 7, Fig. 13.

*M. laticeps* Nordstedt. Duncan Lake. Pl. 7, Fig. 14.

*M. pinnatifida* (Kuetzing) Ralfs. Hematite Lake. Pl. 7, Fig. 15.

*M. truncata* (Corda) Brebisson. Duncan and Hematite lakes. Pl. 8, Fig. 1.

*Sphaerosozma aubertianum* West. Hematite Lake. Pl. 8, Fig. 2.

*Teilingia excavata* (Ralfs) Bourrelly. Duncan and Hematite lakes. Pl. 8, Fig. 3.

*Onychonema laeve* Nordsted. Duncan and Hematite lakes. Pl. 8, Fig. 4.

*Spondylosium moniliforme* Lundell. Duncan and Hematite lakes. Pl. 8, Fig. 5.

*Hyalotheca dissiliens* (J.E. Smith) Brebisson. Duncan, Energy, and Hematite lakes. Pl. 8, Fig. 6.

*H. mucosa* (Mertens) Ehrenberg. Duncan and Hematite lakes. Pl. 8, Fig. 7.

*Desmidium aptogonum* Brebisson. Duncan and Hematite lakes. Pl. 8, Fig. 8.

*D. grevillii* (Kuetzing) DeBary. Hematite Lake. Pl. 8, Fig. 9.



*D. swartzii* Agardh. Duncan and Hematite lakes. Pl. 8, Fig. 10.

*Bambusina brebissonii* Kuetzing. Hematite Lake. Pl. 8, Fig. 11.

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# Effect of Artificial Nest Density and Wetland Size on Canada Goose Clutches in Constructed Wetlands near Cave Run Lake, Kentucky

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## ABSTRACT

Artificial nesting structures have commonly been used to increase the breeding success of Canada geese, *Branta canadensis*, in wetland habitats. We examined the effects of nest density and wetland size on brood size and hatching success in recently constructed wetlands (mean = 10 years old) near Cave Run Lake, Kentucky. Wetland basins were 0.2–16.1 ha; nest densities ranged from 0.2–7.4 structures per hectare. The average clutch was smaller than most previous researchers have found (3.6). The percentage that hatched (62%) was in the normal range. Density did not affect the number of eggs laid ( $r^2 = 0.09$ ,  $P = 0.09$ ,  $n = 33$ ) or the percentage that hatched ( $r^2 = 0.07$ ,  $P = 0.13$ ,  $n = 33$ ). Larger wetlands had larger clutches ( $r^2 = 0.13$ ,  $P = 0.04$ ,  $n = 33$ ), but the percentage of eggs that hatched was not significantly greater ( $r^2 = 0.08$ ,  $P = 0.01$ ,  $n = 33$ ). We surmised that these young constructed wetlands may not yet be providing sufficient food for egg laying, but that larger wetlands can provide better habitat, even at nest densities up to 7/ha. These constructed wetlands with artificial nests do not provide the same resources and habitat to Canada geese as natural wetlands.

## INTRODUCTION

The number of waterfowl is directly related to the amount of suitable wetland habitat in a region (Merendino et al. 1995). Wetland habitat is rare in Daniel Boone National Forest; therefore, 110 wetlands with a combined area of almost 69 ha were constructed by the U.S. Forest Service to enhance populations of wetland flora and fauna. As part of this effort, artificial nesting structures were placed in these wetlands in an attempt to increase breeding success of Canada geese (*Branta canadensis*).

Artificial structures have been found to produce more goslings than natural shoreline nesting sites (Ball 1990). When these nests are placed in wetlands at higher densities than the habitat can support, density dependent theory suggests that some factor should cause population growth to decline. One way birds can accomplish this is to reduce reproductive output—for example by laying fewer eggs or by nest abandonment. High nest densities can result in nest desertion due to aggressive conflicts between neighboring breeding pairs (Ewaschuk and Boag 1972; Lokemoen and Woodward 1992).

The size of the wetland also affects reproductive success. Larger wetlands should have more loafing sites for ganders and more available food. The objective of our research was to determine if nest density and wetland size

affect two factors of Canada goose reproductive success: clutch size and the percentage of eggs that hatch.

## MATERIALS AND METHODS

The 33 wetlands assessed are located in northeastern Kentucky in Daniel Boone National Forest. The watershed of the study area is dominated by mixed-mesophytic forest in sparsely populated regions of Rowan, Bath, Menifee, and Rowan counties near Cave Run Lake.

All the constructed wetlands were relatively young: some were built 24 years before our study, some less than 2 years (mean =  $9.7 \pm 8.6$ ;  $\pm$  SE). The wetlands were built by digging out and sometimes diking suitable sites. All had some water control structure to allow filling and draining. Wetland sizes ranged from 0.2 to 16.2 ha. Most wetlands were 0.4 to 2.4 ha; only three were larger than 3 ha. All the wetlands were roughly rectangular and about 1–2 meters deep. There was little variation in shape or water depth among sites. We assessed 56 artificial nests placed in open water. Artificial nest densities ranged from 0.2 to 7.4 structures/ha (mean =  $2.1 \pm 1.6$ ). Emergent vegetation never covered more than 30% of any wetland (mean = 6%). The landscape around each basin was similar: forest and open water. The amount of forest directly adjacent



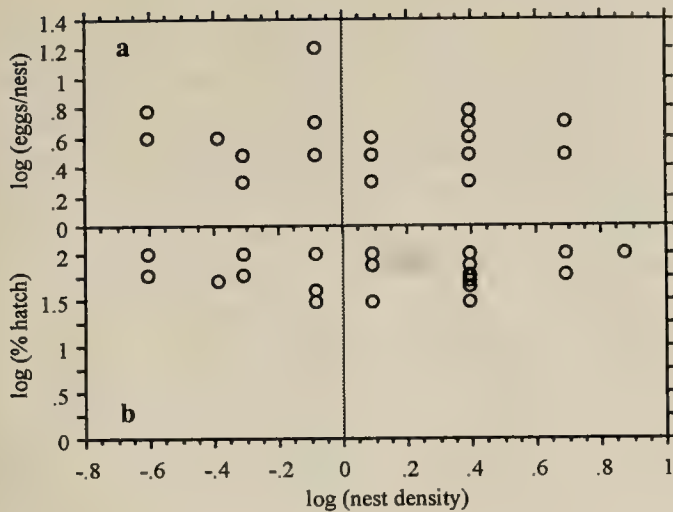


Figure 1. Relationship between artificial nest density (nest/ha) and clutch size (a) and hatching success (b) among Canada geese in constructed wetlands near Cave Run Lake, Kentucky.

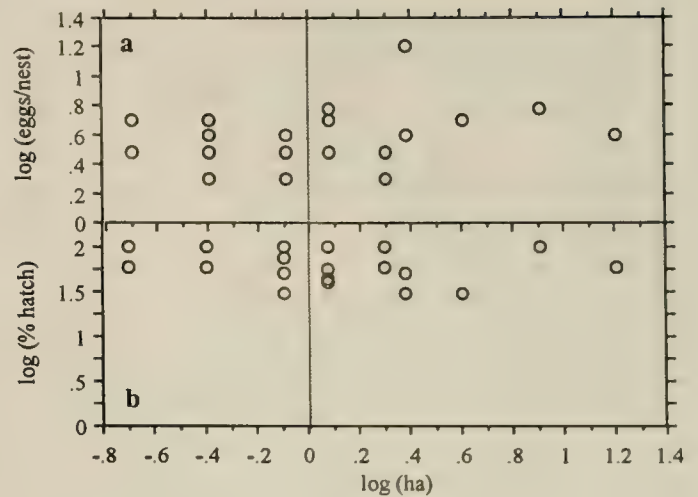


Figure 2. Relationship between wetland size (ha) and clutch size (a) and hatching success (b) among Canada geese in constructed wetlands near Cave Run Lake, Kentucky.

to the wetland had no significant affect on reproductive success.

Field data were collected from 11 Mar–5 May 1996. Each nesting structure was observed, from cover, for 30 minutes at least six times. Clutch counts (number of eggs/nest) were obtained 23–27 Apr 1996; egg survival (percentage of eggs that successfully hatched) was surveyed 17–23 May 1996. Normal biases in calculating egg success (Mayfield 1975) were not applicable because success was not measured as a minimum of one hatch per nest and no nests were abandoned or destroyed.

Because the data included counts, densities, and percentages, they were log transformed to fit the assumptions of parametric statistical models (Zar 1984). Effects of density-dependence and habitat size are usually assumed to be linear (Colinvaux 1993); therefore, we used a simple linear regression model, with nest density and wetland size as independent variables, and eggs/nest and hatching success as dependent variables. Analysis of variance testing was used to determine if regressions were significant (Zar 1984). We chose 95% confidence as our significance limit for all analyses.

## RESULTS

All structures studied were used by Canada geese. Out of the 226 eggs laid during the breeding season, 62% produced live goslings. The average clutch had  $3.6 \pm 2.6$  eggs. Two structures were used by more than one mating pair at the same time (gang nesting). No eggs

hatched from those nests. All other nests had at least one egg hatch (93.9% nest success).

Geese nesting closer together laid fewer eggs, but the effect of density was insignificant ( $r^2 = 0.09$ ,  $P = 0.09$ ,  $n = 33$ , Figure 1a). The percentage of eggs that hatched was not significantly influenced by nest density ( $r^2 = 0.07$ ,  $P = 0.13$ ,  $n = 33$ , Figure 1b). Larger wetlands had larger clutches ( $r^2 = 0.13$ ,  $P = 0.04$ ,  $n = 33$ , Figure 2a). Larger wetlands also seemed to have a greater chance of having a successful hatch, but the effect was insignificant ( $r^2 = 0.08$ ,  $P = 0.10$ ,  $n = 33$ , Figure 2b).

## DISCUSSION

Our geese laid fewer eggs per nest than most other researchers have found, but our hatching rates are within the range of other studies (3.6 eggs/nest with 63% hatching). Geis (1956) found a mean clutch size of 5.4, with 2.9 eggs hatching (54%). Data summarized by Lebeda and Ratti (1983) indicated an average clutch size of 4.4 and 62% brood success—with 1 or more eggs hatching from 56% of all nests observed. Brakhage (1965) compiled data from previous studies and found an average clutch of 5.1, with 73–93% successfully hatching. Brakhage (1965) studied Canada geese in artificial nests at high densities (about 60 meters between nests). He reported open-water nesters had an average clutch of 5.5 eggs with 72% hatching.

Although our clutches were smaller than normal, we did not find a significant effect of



high nest density on the number of eggs laid. This could also be attributed to the nest being in the open water (Gosser and Conover [1999] found that geese prefer islands to shoreline edges), in a relatively undisturbed surrounding landscape. Ewaschuk and Boag (1972) and Kossack (1950) found that high nest densities result in greater numbers of agonistic interactions between nesting pairs, resulting in high desertion rates. Ewaschuk and Boag (1972) described a correlation between density and nest success, but only five data points were used to determine the relationship. The lake island they studied had densities ranging from an extraordinary 20–23 nests/ha (an order of magnitude greater than our average density). They found some of the lowest success rates compared to other studies (averaging 52% of nest hatching one or more eggs), which they attributed mostly to agonistic interactions and predation. Gloutney et al. (1993) found that human disturbance influences nest fate, especially in the early stages of egg laying.

Agonistic interactions, predation, and human disturbance were not significant in our study; no nests were deserted. This was probably because all of our wetlands are in low population areas surrounded by intact forests. Therefore, some other factor must have reduced the brood size. Young geese typically have smaller clutches (Brakhage 1965); so a possible explanation is the geese we examined were younger.

A more plausible explanation is that the geese could not obtain sufficient food (Martin 1987). All wetlands had some forest nearby providing adequate cover, but they may not have had sufficient forage. Dense growth of emergent vegetation can increase nesting success (Ewaschuk and Boag 1972; Poly 1979). Our sites averaged 6% emergent vegetative cover. Recently constructed ecosystems are not providing the functions and values of natural habitats (McKinstry and Anderson 1994; Weller 1990). Further support of this theory is that larger wetlands (presumably with more resources) had larger broods and somewhat higher hatching rates.

We found a significant effect of island size on clutch size. Geis (1956) noted that large islands (>10 ha) had an order of magnitude more nests per unit area than small (<0.5 ha)

islands. Geis's highest nest densities were only about 1.2 nest/ha on Flathead Lake, Montana. High densities can result in more gang brooding (Brakhage 1965), but Warhurst and Bookhout (1983) found gang brooding did not affect reproductive success in diked Lake Erie marshes when densities were 3.1 nest/ha.

We can conclude that nests can be placed in high densities in wetlands without having a significant effect on hatching success. To increase breeding success of Canada geese, managers should construct wetlands as large as possible, and in relatively undisturbed areas. Building fewer large wetlands could have a negative effect on biodiversity. Brown and Dinsmore (1986) found that wetland complexes with many small wetlands supported a higher diversity of waterfowl than a single large area. Landscape position should also affect success. Sites with nearby sources of food would be favorable. We did not examine surrounding landscape in detail because of our lack of surrounding land use diversity. We did find it is important to encourage the growth of emergent aquatic macrophytes to provide a suitable habitat and food.

#### ACKNOWLEDGMENTS

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# Surveys of Bird Communities on Little Black and Black Mountains: Implications for Long-term Conservation of Montane Birds in Kentucky

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## ABSTRACT

Considerable attention has been placed on cataloging and protecting the avifaunal communities inhabiting forests at the highest elevations (circa. 1200 m and above) in the Cumberland Mountains of southeastern Kentucky. However, few data are available on forest bird communities at lower elevations in this region, particularly locations historically or presently subject to disturbance from logging or mineral extraction. I surveyed bird communities at 16 forested sites in May and June 1999 at Little Black and Black mountains at elevations ranging from 730 to 1005 m. Sites surveyed were historically disturbed by logging and or mining and were proposed for additional logging and mining activities. Surveys indicated that bird communities were a mix of forest interior and early-successional species and included birds of both northern and southern affinities. Nine species known from the highest elevations in Kentucky were present. Species recognized by the Kentucky State Nature Preserves Commission as having special concern status in the state that were recorded during surveys included Blackburnian warbler (*Dendroica fusca*), Canada warbler (*Wilsonia canadensis*), cerulean warbler (*D. cerulea*), dark-eyed junco (*Junco hyemalis*), golden-winged warbler (*Vermivora chrysoptera*), and rose-breasted grosbeak (*Pheucticus ludovicianus*). These data suggest that habitat exists at mid-elevations in the Cumberland Mountains for several rare species of birds in Kentucky. A long-term strategy for monitoring the effects of land-use practices on bird communities in the Cumberland Mountains is recommended.

## INTRODUCTION

The status of forest-dependent bird species has become a topic of increasing attention, as land-use practices continue to alter the landscape pattern in heavily-forested regions (Askins et al. 1990; Freemark and Collins 1992; Robbins et al. 1989; Rosenberg et al. 1999; Walters 1998). Conversion of contiguous forest into a fragmented mosaic of forest and non-forest habitats is believed to alter population levels and bird species composition (Faaborg et al. 1995; Robinson and Robinson 1999; Robinson and Wilcove 1994). It has been hypothesized that a reduction in block size of forest habitats can lead to source-sink dynamics, such that smaller blocks provide insufficient conditions for adequate replacement through reproduction in existing breeding bird communities (Rosenberg et al. 1999; Schmidt and Whelan 1999). Loss of available forest habitat notwithstanding, declines in abundance and reproductive performance of breeding birds have been attributed to brood parasitism by brown-headed cowbirds, *Molothrus ater* (Brittingham and Temple 1983; Mayfield 1977; Robinson, Rothstein et al. 1995), nest predation by vertebrate predators

(Martin 1992; Robinson 1992; Wilcove 1985), and loss of basal area and changes in forest structure (Annand and Thompson 1997; Baker and Lacki 1997; Buford and Capen 1999; Robinson and Robinson 1999; Schulte and Niemi 1998).

The Cumberland Mountains in southeastern Kentucky support a predominantly forested landscape that provides habitat for a rich mix of bird species, including many with northern and southern affinities (Mengel 1965; Palmer-Ball 1996). Included within this region are forest habitats situated at the highest elevations in the state, on mountain tops achieving elevations in excess of 1200 m (Fenneman 1938). These high-elevation habitats support a number of bird species believed to breed nowhere else in Kentucky (Mengel 1965; Palmer-Ball 1996). Although both breeding and non-breeding surveys of birds exist for several locations in the Cumberland Mountains (e.g., Croft 1969; Davis et al. 1980; Wetmore 1940), most attention has been paid to bird communities of Black Mountain, with disproportionate sampling effort allocated to the highest elevations (Barbour 1941; Breiding 1947; Davis and Barbour 1978; Davis and



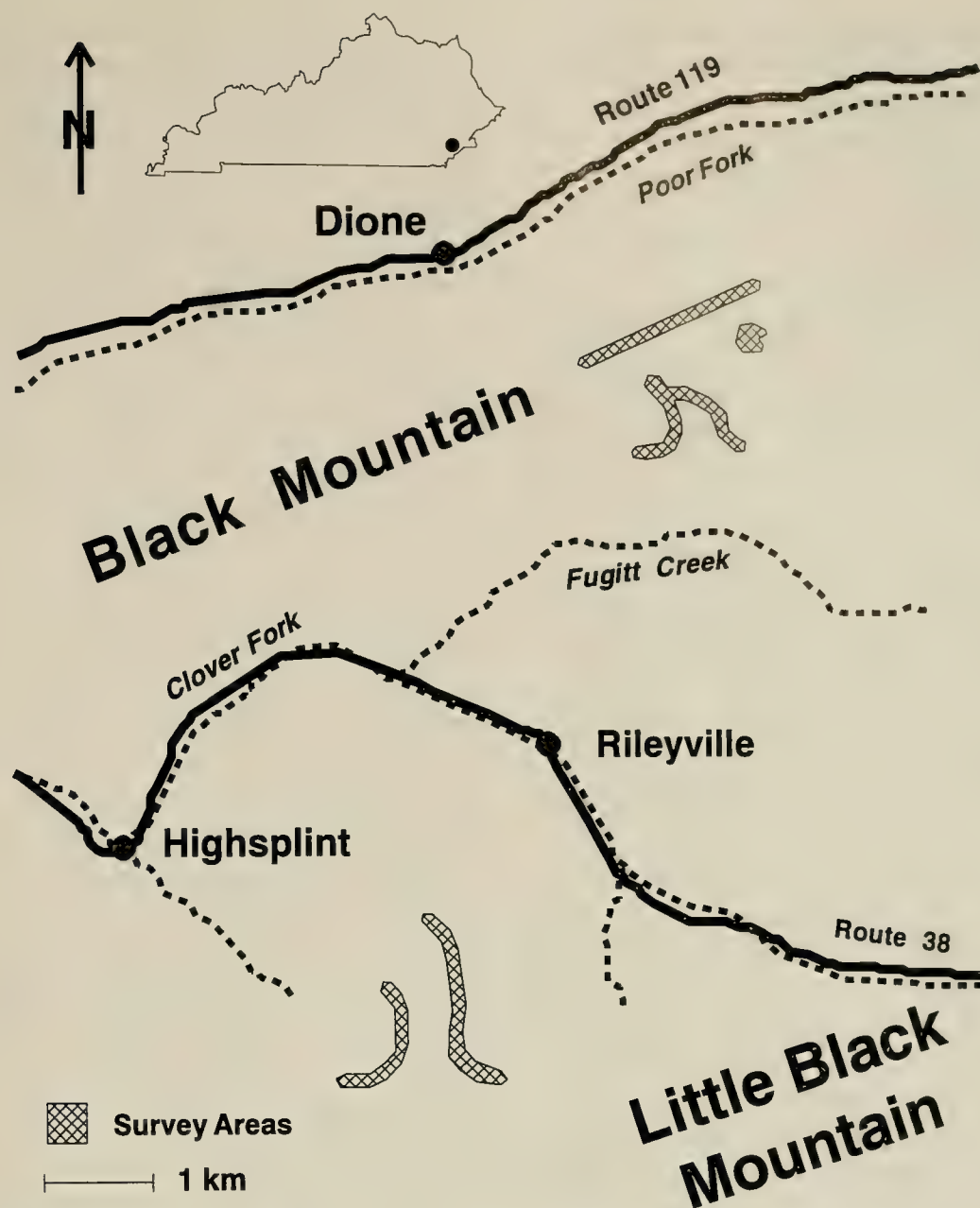


Figure 1. Location of areas where birds were surveyed on Little Black and Black mountains, Harlan County, Kentucky, in May and June 1999. Inset: Outline map of Kentucky showing location of survey area.

Smith 1978; Howell 1910; Lovell 1950a, 1950b). Much less is known about breeding bird communities at mid-elevations and at lower summits, such as on Little Black Mountain.

Data quantifying the response of bird communities to changes in forest habitats in Kentucky are limited (Baker 1996; Baker and Lacki 1997; McComb and Muller 1983; McComb et al. 1991), with virtually no data existing for the Cumberland Mountains. In this paper, I present baseline data on bird communities and existing habitat conditions in forest habitats at Little Black and Black mountains, where a history of logging and or mining disturbance exists and where future distur-

bance events are planned. The results of these surveys are compared to historical patterns of occurrence and abundance of bird species in the Cumberland Mountain region; the implications for long-term conservation of montane bird communities in Kentucky are discussed.

#### STUDY AREA AND METHODS

Survey sites were located in Harlan County, Kentucky, at Little Black and Black mountains (Figure 1). Sites included both northwest and southeast facing slopes at elevations ranging from 730 to 1005 m on both mountains. Little Black and Black mountains are situated in the Cumberland Mountains physiographic section (Fenneman 1938), a segment of the state com-



prising the Cumberland Crest avifaunal region (Mengel 1965). The Cumberland Mountains comprise 2% of the land mass in Kentucky and contain the highest elevations in the state, reaching a maximum on Black Mountain of 1265 m (Palmer-Ball 1996). Little Black Mountain lies to the southeast of Black Mountain, with a summit lower (ca. 1100 m) in elevation. The mountains represent a syncline with horizontal strata comprised of shales, coal, and sandstone of the Pennsylvanian series (Braun 1940; Lovell 1950b). Soils are largely mature with deep horizons (Braun 1940).

The vegetation of the Black Mountain region is composed of a matrix of segregates of the mixed mesophytic forest association (Braun 1940). Historically, oak-chestnut forest predominated on drier slopes and ridges (Braun 1940); however, American chestnut (*Castanea dentata*) has largely disappeared, as evidenced by Lovell (1950b) describing "skeletons of many large chestnuts" at Black Mountain in the late 1940s. The composition of vegetation in the Black Mountain area differs from that of adjacent mountains by exhibiting a lack of red spruce (*Picea rubens*) and fraser fir (*Abies fraseri*) trees, and having a limited abundance of pines (*Pinus virginiana*) (Braun 1940; Croft 1969; Lovell 1950b).

The Cumberland Mountains were primarily forested prior to settlement, with open habitat limited to cliffs, rock outcrops, riparian zones, and natural disturbances such as fire and windthrow (Palmer-Ball 1996). The region remains forested, especially at higher elevations, but alteration of some of the landscape has occurred due to logging, mining, agricultural uses, and land clearing for settlements (Palmer-Ball 1996). The original mixed mesophytic forest does remain at higher elevations where logging, fire, or mining have not occurred (Palmer-Ball 1996).

Birds were surveyed using a modified, fixed-radius point-count method (Blondel et al. 1970; Hutto et al. 1986). Bird occurrence, primarily of singing males, was recorded by species in concentric distance bands of <50 m and  $\geq 50$  m (Ralph et al. 1993). Surveys were made at 16 survey sites, eight each at Little Black and Black mountains. All birds seen or heard during surveys were recorded. All surveys were conducted between 0600 and 0900

EDST to coincide with peak singing activity of the majority of bird species (Robbins 1981). Survey periods were 12 min in duration, waiting 3 min from the time of arrival at the survey site prior to beginning data collection to allow disturbed birds to resume normal singing activity (Reynolds et al. 1980). Surveys were conducted on 12 and 13 May and 15 and 16 June 1999 at Little Black Mountain, and on 14 and 15 May and 17 and 18 June at Black Mountain. All survey sites were sampled once in May and once in June. Sampling in both May and June accounts for the use of the site by migrants and early and late-breeding residents (Baker and Lacki 1997). Weather was favorable for all surveys, with temperatures ranging from 52° to 66°F. Light rain occurred periodically on the morning of 13 May but did not appear to affect surveys as birds continued to sing throughout the drizzle.

A qualitative description of the vegetation was made at each survey site in June. A list of species present in the mid- and overstory was compiled with the dominant and co-dominant tree species recorded. Stem diameters of the dominant species at each survey site were categorized as follows: <30 cm, 30–45 cm, >45–60 cm, and >60 cm. Further, evidence of disturbance was noted and successional status at each survey site categorized as recently logged, early successional, mid-successional, or intact second-growth forest.

Data on bird occurrence within 50 m from the observation point at survey sites were compared between locations and months of sampling by species richness (i.e., number of species,  $S$ ), bird abundance (i.e., number of birds,  $N$ ), and species diversity (i.e., Shannon index,  $H'$ ); the latter was calculated using base10 logarithms with the program provided in Ludwig and Reynolds (1988). Data were analyzed by two-way analysis of variance, with an alpha of 0.05 as the level of significance.

## RESULTS

There was no difference between survey sites at Little Black Mountain and Black Mountain for either species richness ( $F = 0.49$ ,  $P = 0.49$ ), bird abundance ( $F = 0.10$ ,  $P = 0.76$ ), or species diversity ( $F = 0.64$ ,  $P = 0.43$ ); however, differences were observed among months of sampling for species richness ( $F = 4.37$ ,  $P = 0.046$ ) and bird abun-



Table 1. Mean  $\pm$  SE for species richness (number of species, S), bird abundance (number of birds, N), and species diversity (Shannon index, H') of bird communities surveyed on Little Black and Black mountains, Kentucky, in May and June 1999. Data are based on eight surveys per location  $\times$  month combination and include only birds recorded within 50 m of sampling points.

Month	Parameter	Little Black Mountain	Black Mountain
May	S	12.2 $\pm$ 1.24	11.0 $\pm$ 1.20
	N	17.5 $\pm$ 2.29	17.8 $\pm$ 1.73
	H'	2.38 $\pm$ 0.11	2.26 $\pm$ 0.12
June	S	9.50 $\pm$ 0.89	9.25 $\pm$ 1.01
	N	14.1 $\pm$ 1.63	12.8 $\pm$ 1.50
	H'	2.15 $\pm$ 0.11	2.08 $\pm$ 0.15

dance ( $F = 5.49$ ,  $P = 0.03$ ), with species richness and bird abundance higher in May than in June (Table 1). No difference was observed between months of sampling for species diversity ( $F = 3.09$ ,  $P = 0.09$ ). All interactions between location and month of sampling were not significant ( $P > 0.05$ ).

I detected 44 species of birds within 50 m of sampling points and recorded an additional seven species either beyond 50 m or while in transit among survey sites (Table 2). All total, I observed 47 species at Little Black Mountain and 39 species at Black Mountain. Birds observed only at Little Black Mountain included American crow (*Corvus brachyrhynchos*), Blackburnian warbler (*Dendroica fusca*), common yellowthroat (*Geothlypis trichas*), dark-eyed junco (*Junco hyemalis*), eastern phoebe (*Sayornis phoebe*), mourning dove (*Zenaida macroura*), northern cardinal (*Cardinalis cardinalis*), ovenbird (*Seiurus aurocapillus*), ruby-throated hummingbird (*Archilocus colubris*), yellow-bellied sapsucker (*Sphyrapicus varius*), yellow-billed cuckoo (*Coccyzus americanus*), and yellow-breasted chat (*Icteria virens*). Birds observed only at Black Mountain included orchard oriole (*Icterus spurius*), white-eyed vireo (*Vireo griseus*), and yellow-rumped warbler (*D. coronata*). Species of birds observed that are recognized as endangered (E), threatened (T), or of special concern (S) status in Kentucky (KSNPC 1996), included Blackburnian warbler (T), Canada warbler (*Wilsonia canadensis*, S), cerulean warbler (*D. cerulea*, S), dark-eyed junco (S), golden-winged warbler (*Vermivora chrysoptera*, T), and rose-breasted grosbeak (*Pheucticus ludovicianus*, S).

Table 2. Species and numbers of birds recorded within 50 m of sampling points at survey sites on Little Black and Black Mountains, Kentucky, in May and June 1999. Additional species recorded beyond 50 m or observed in transit among survey sites are considered anecdotal and are indicated by an (\*).

Species	Little Black Mountain		Black Mountain	
	May	June	May	June
<i>Archilochus colubris</i>		1		
<i>Bonasa umbellus</i>	*		*	
<i>Cardinalis cardinalis</i>	6	4		
<i>Carduelis tristis</i>	3	1	3	2
<i>Catharus fuscescens</i>	1	2	4	1
<i>Coccyzus americanus</i>		*		
<i>Colaptes auratus</i>		*	3	
<i>Contopus virens</i>		*	1	2
<i>Corvus brachyrhynchos</i>	*			
<i>Cyanocitta cristata</i>	1	2	*	3
<i>Dendroica caerulescens</i>	9	3	10	8
<i>D. cerulea</i>	2	2	2	
<i>D. coronata</i>			1	
<i>D. fusca</i>		1		
<i>D. pensylvanica</i>	8	10	12	5
<i>D. tigrina</i>	2		1	
<i>D. virens</i>	2	3		
<i>Dryocopus pileatus</i>		*	1	1
<i>Empidonax virescens</i>	1			2
<i>Geothlypis trichas</i>	1			
<i>Helmitheros vermivorus</i>	1	1		1
<i>Hylocichla mustelina</i>	9	2	10	4
<i>Icteria virens</i>	1			
<i>Icterus spurius</i>			1	
<i>Junco hyemalis</i>		*		
<i>Meleagris gallopavo</i>	*		*	
<i>Mniotilta varia</i>	8	4	7	3
<i>Ophorornis formosus</i>	4	4	1	*
<i>Parus bicolor</i>	3	1	*	1
<i>P. carolinensis</i>		5	4	*
<i>Passerina cyanea</i>	8	7	4	2
<i>Pheucticus ludovicianus</i>	5	4	7	2
<i>Picoides pubescens</i>	2	1	1	1
<i>P. villosus</i>			1	1
<i>Pipilo erythrophthalmus</i>	10	4	11	6
<i>Piranga olivacea</i>	5	5	2	6
<i>Polioptila caerulea</i>		3		4
<i>Sayornis phoebe</i>	*			
<i>Seiurus aurocapillus</i>	3		5	4
<i>Setophaga ruticilla</i>	8	7	4	5
<i>Sitta carolinensis</i>		1	2	7
<i>Sphyrapicus varius</i>	1			
<i>Thryothorus ludovicianus</i>	4	1	3	1
<i>Vermivora chrysoptera</i>	*		1	
<i>Vireo flavifrons</i>	2		*	
<i>V. griseus</i>			*	
<i>V. olivaceus</i>	10	6	16	11
<i>V. solitarius</i>	1	7	4	5
<i>Wilsonia canadensis</i>	3	1		*
<i>W. citrina</i>	16	16	20	14
<i>Zenaida macroura</i>		1		



Table 3. Number of survey sites with woody plant species at Little Black and Black mountains, Kentucky, in June 1999. Number of sites where the species was identified as a canopy dominant is indicated in parentheses. Data are based on eight sites per location.

Species	Number of sites	
	Little Black Mountain	Black Mountain
<i>Acer pensylvanicum</i>	1	
<i>A. rubrum</i>	3	5 (4)
<i>A. saccharum</i>	5 (1)	3 (2)
<i>Amelanchier laevis</i>		1
<i>Betula lenta</i>	5 (1)	
<i>Carya</i> sp. (rough-bark)	1	
<i>Carya</i> spp. (smooth-bark)	1	3
<i>Fraxinus americana</i>	3	3
<i>Liriodendron tulipifera</i>	8 (3)	6 (4)
<i>Magnolia acuminata</i>	2	1
<i>Morus rubra</i>		1
<i>Nyssa sylvatica</i>		1
<i>Oxydendrum arboreum</i>		2
<i>Pinus virginiana</i>		1 (1)
<i>Prunus serotina</i>	2	2
<i>Quercus alba</i>		2
<i>Q. falcata</i>	1	
<i>Q. muehlenbergii</i>	3 (1)	3
<i>Q. prinus</i>		3 (2)
<i>Q. rubra</i>	2	3 (1)
<i>Robinia pseudoacacia</i>	8 (7)	5 (1)
<i>Salix nigra</i>	2	
<i>Sassafras albidum</i>	1	2
<i>Tilia</i> spp.		4 (2)

Species recorded most frequently within 50 m of survey points, included black-throated blue warbler (*D. caerulescens*, N = 30), chestnut-sided warbler (*Dendroica pensylvanica*, N = 35), hooded warbler (*Wilsonia citrina*, N = 66), red-eyed vireo (*Vireo olivaceus*, N = 43), and rufous-sided towhee (*Pipilo erythrophthalmus*, N = 31). I observed no brown-headed cowbird (*Molothrus ater*), a brood parasite, but did record two avian nest predators, American crow and blue jay (*Cyanocitta cristata*).

I recorded 24 species of trees at survey sites (Table 3). Black locust (*Robinia pseudoacacia*), sugar maple (*Acer saccharum*), sweet birch (*Betula lenta*), and yellow-poplar (*Liriodendron tulipifera*) were most frequent at Little Black Mountain; basswood (*Tilia* sp.), black locust, red maple (*A. rubrum*), and yellow-poplar were most frequent at Black Mountain. Maximum diameter size class at survey sites at Little Black Mountain ranged from <30 cm to 60cm, and from <30 cm to >60 cm at

Table 4. Number of survey sites with a maximum diameter size class of canopy trees and stage of succession at Little Black and Black mountains, Kentucky, in June 1999. Data are based on eight sites per location.

	Number of sites	
	Little Black Mountain	Black Mountain
Maximum diameter class		
<30 cm	3	2
30–45 cm	3	0
>45–60 cm	2	4
>60 cm	0	2
Stage of succession		
Recently logged	4	0
Early successional	4	3
Mid-successional	0	3
Intact second-growth forest	0	2

Black Mountain (Table 4). All survey sites at Little Black Mountain were either in an early successional stage or were logged just prior to surveys. Habitats at survey sites at Black Mountain were more varied and included early, mid-, and late successional stages.

DISCUSSION

After surveying birds at four major mountain groups in southeastern Kentucky, Croft (1969) concluded that the higher elevations in Kentucky were characterized by a meager “complement of northern species.” He attributed his observations to the absence or paucity of coniferous tree species and to the harmful effects of logging and mining in the region. Further, he suggested that mined lands supported an overall lower abundance of birds when compared with habitats of undisturbed, intact forest at the same elevation. My survey results paralleled his observations. Mean values for species richness, bird abundance, and species diversity were consistently below values reported by Baker and Lacki (1997) for bird communities on the Cumberland Plateau, Kentucky. Moreover, mean values for bird communities on Little Black and Black mountains were lower regardless of whether the data were compared to bird communities in no-harvest, high-leave harvest, low-leave harvest, or clearcut harvest sites (Baker and Lacki 1997); however, sites sampled by Baker and Lacki (1997) were at elevations below 400 m. Given that many of the sites surveyed in this study were already im-



pacted by logging or mining to some extent, attributing lower mean values of richness, abundance, and diversity of birds to disturbance effects or to the shorter growing season at higher elevations cannot be resolved. The differences observed between months of sampling for species richness and bird abundance were likely due to the presence of migrants being recorded as they were passing through in May. Similar trends have been reported elsewhere in the state (Baker and Lacki 1997; Lacki and Baker 1998).

Data for tree species composition at survey sites did reveal a limited frequency of conifer species, as predicted by Braun (1940), Croft (1969), and Lovell (1950b). Virginia pine (*Pinus virginiana*), the only conifer recorded, was found at only a single survey site on Black Mountain. A lack of conifers likely prevents summer residence by some species that breed in coniferous forests further south in latitude at high elevations, such as golden-crowned kinglet (*Regulus satrapa*), red-breasted nuthatch (*Sitta canadensis*), and winter wren (*Troglodytes troglodytes*) (Alsop 1991; Rabenold et al. 1998). Thus, these data concur with those of Croft (1969), and suggest that the paucity of coniferous tree species serves as an additional constraint on the richness, abundance, and diversity of birds in the Cumberland Mountain region.

Mengel (1965) labeled the bird community of the Cumberland Mountains as the Cumberland Crest Avifaunal region and attributed its distinctiveness to the presence of nine bird species that do not breed elsewhere in Kentucky: blackburnian warbler, black-throated blue warbler, blue-headed vireo (*Vireo solitarius*), Canada warbler, chestnut-sided warbler, dark-eyed junco, golden-winged warbler, rose-breasted grosbeak, and veery (*Catharus fuscescens*). Recent studies have demonstrated that the blue-headed vireo breeds elsewhere in the state (Lacki and Baker 1998; Yacek and Lacki 1998). Regardless, all nine species were observed in this study, suggesting that forests at mid-elevations at Little Black and Black mountains support breeding habitat for these rare species of birds. Of these, only golden-winged warbler was not observed in June, the period in which singing males are reflective of breeding residents. Croft (1969) saw this spe-

cies on a number of occasions in June though, at both Pine and Black mountains.

Golden-winged warblers are known to hybridize with blue-winged warblers (*Vermivora pinus*) in Kentucky (Palmer-Ball 1996); Croft (1969) reported on a golden-winged warbler emitting the song of a blue-winged warbler during his surveys. The bird was situated at an elevation of 670 m in the valley between Pine and Black mountains. I also observed a golden-winged warbler singing a song intermediate between its typical song and that of a blue-winged warbler. This bird was observed shortly after I completed a survey on 13 May at an elevation of 760 m at Little Black Mountain. The bird was perched in a willow (*Salix nigra*) thicket singing a “bee buzz buzz” song, one “buzz” note short of the typical golden-winged warbler song and one “buzz” note longer than that of the blue-winged warbler song. I observed two other golden-winged warblers, but none was heard singing. Although common elsewhere in the state (Palmer-Ball 1996), blue-winged warblers have yet to be reported in surveys of the Cumberland Mountains (Croft 1969; Davis et al. 1980; Mengel 1965; this study); thus, the status of the blue-winged  $\times$  golden-winged warbler complex in the Cumberland Mountains remains unresolved.

Based on the summarized historic and atlas survey data in Palmer-Ball (1996), no prior record of a dark-eyed junco exists for Little Black Mountain, extending the known summer range of this species in the state. This bird was observed at ca. 0800 on the morning of 15 June while in transit between survey sites. The bird was perched within 5 m of the intersection of two logging roads at the crest of the mountain. An immature orchard oriole was recorded during surveys at Black Mountain on 14 May. This bird was perched at the edge of a small clearing (<0.5 ha in size) in otherwise intact, second growth forest at an elevation of 850 m. This species is known to occur at lower elevations in the Cumberland Mountains (Croft 1969; Howell 1910) and was classified as a probable breeding resident in Harlan County, Kentucky (Palmer-Ball 1996). This observation would suggest that the species does breed in Harlan County and occurs at elevations somewhat higher than previously reported.

Although hooded warblers and red-eyed vir-



eos were the two most frequently observed species, data indicated that several edge or shrub-scrub species were prevalent at survey sites on Little Black or Black mountains, including American goldfinch (*Carduelis tristis*), Carolina wren (*Thryothorus ludovicianus*), chestnut-sided warbler, indigo bunting (*Passerina cyanea*), northern cardinal, and rufous-sided towhee. These observations reflect the successional status of vegetation at most survey sites and are consistent with data for tree species composition that demonstrated dominance by invasive species such as black locust, yellow-poplar, and red maple. Croft (1969) hypothesized that mining in the Black Mountain region was likely resulting in a lower abundance of forest-inhabiting birds and leading to an increase in disturbance associated species. These data indicate that shrub-scrub species have likely benefited from the increase in edge habitat; however, the abundance of American redstart (*Setophaga ruticilla*), black-and-white warbler (*Mniotilta varia*), black-throated blue warbler, hooded warbler, rose-breasted grosbeak, and wood thrush (*Hylocichla mustelina*) observed in this study indicate that forest-inhabiting birds will recolonize previously disturbed sites as succession leads to an increase in vegetative complexity and stratification (Balda 1975; Niemi and Hanowski 1984). In fact, Croft's (1969) suggestion that the hooded warbler would likely be the species most impacted by strip mining is not supported by these data.

Point count data do not directly reflect breeding success of singing males because the most critical element of breeding habitat is nest site quality (Merrill et al. 1998). For example, birds heard during surveys may represent unmated males in residual patches of habitat (Gibbs and Faaborg 1990; Probst and Hayes 1987; Van Horn et al. 1995) or immigrants drawn to sink habitats from source populations elsewhere (Pulliam 1988; Martin 1992; Robinson 1992). Thus, any inferences drawn solely from point count data must be viewed with caution. The habitat measures obtained, however, indicate that breeding habitat of forest-inhabiting birds at mid-elevations in the Cumberland Mountains continues to be altered by logging and mining activities and is consistent with reported findings (Croft 1969; McComb et al. 1991; Mengel 1965). The

dominant vegetation characteristic of mixed mesophytic forest (Braun 1940), has been replaced in many localities by direct plantings of black locust, along with volunteer species typical of disturbed habitats, such as yellow-poplar and sassafras (*Sassafras albidum*) (Croft 1969; McComb et al. 1991; this study). Thus it is likely that populations of forest-inhabiting birds are being negatively impacted both by fragmentation, particularly where mined lands are returned to fescue (*Festuca arundinacea*) grasslands, and by changes in the condition of existing forest habitat (McComb et al. 1991).

Of added concern are the potential effects that nest parasitism by the brown-headed cowbird and nest predation by vertebrate predators could have on breeding success of forest-inhabiting birds of the Cumberland Mountains. Fragmentation of forests, regardless of whether it is by logging, mining, or agriculture, increases edge habitat and is known to be associated with increased levels of nest parasitism and nest predation (Brittingham and Temple 1983; Martin 1988; Robinson, Thompson et al. 1995; Wilcove 1985). Although no study of nest parasitism exists for the Cumberland Mountain region, brown-headed cowbirds are known to be attracted to reclaimed strip mines in eastern Kentucky (Claus et al. 1988) and have been observed entering larger blocks of interior forest on the Cumberland Plateau (Lacki and Baker 1998). Even though I observed no brown-headed cowbird in this study, Croft (1969) reported this species to be "widespread in moderate numbers" on the ridgetops he surveyed; thus, the potential threat of brood parasitism exists for the Cumberland Mountain region. Rosenberg et al. (1999) have demonstrated that the brown-headed cowbird is less likely to inhabit fragmented habitats at higher elevations throughout the species' distribution.

I observed two avian nest predators during my surveys: American crow and blue jay. These species have also been seen at the higher elevations in the Cumberland Mountains (Breiding 1947; Croft 1969; Wetmore 1940), but their impact on nesting success of breeding birds in Kentucky is unknown. As with the brown-headed cowbird, avian nest predators appear to be less common in fragmented habitats at higher elevations (Rosenberg et al. 1999).



## SUMMARY

McComb et al. (1991) urged for a “proactive plan” to preserve mature forest species of wildlife in the Cumberland Mountains. To reduce the effects of logging and mining they recommended four steps be taken: reforestation of mined lands, carefully planned timber harvests (i.e., location and size), extending the length of timber rotations beyond 80 yrs in some portions of the landscape, and developing a regional plan to optimally allocate the distribution of seral stages. Their overall goal was to minimize cumulative effects of land use on wildlife. Clearly, their message has yet to be heard.

Existing conservation efforts in the Cumberland Mountains of Kentucky have centered around the protection of the highest elevations at the top of Black Mountain and the control of mountaintop removal as a method of coal extraction (KSNPC 1998). Mountaintop removal involves the exposure of coal seams by removal of all biological and mineral material above the surface of the coal. This material is then disposed of in lower lying areas following completion of mining, resulting in permanent disfigurement of the landscape. Although laudable achievements in their own right, limiting the scope of conservation efforts in eastern Kentucky to these two measures has short-sighted and long-term consequences for the bird communities of the Cumberland Mountains. Based on earlier surveys (Croft 1969; Davis et al. 1980; Mengel 1965; Wetmore 1940) and data collected in this study, it is clear that mid-elevations in the Cumberland Mountains support a number of sensitive bird species, including species unique to this region of the state. Protection of only the highest elevations on Black Mountain will provide for a limited acreage of mature forest habitat that could likely be inadequate to sustain breeding populations of these sensitive species over time (Faaborg et al. 1995; Freemark and Collins 1992; Robinson, Thompson et al. 1995; Robinson and Wilcove 1999). Further, even if mountaintop removal is eliminated as a means of coal extraction, unregulated mining at mid-elevations by alternative means could still reduce the amount of available mature forest habitat to where the Cumberland Mountains in Kentucky revert from being a “source” hab-

itat region to that of a “sink” habitat region for forest-inhabiting birds (Pulliam 1988; Rosenberg et al. 1999; Schmidt and Whelan 1999).

The development of a regional plan, as suggested by McComb et al. (1991), could have long-reaching benefits for the conservation of avifauna in eastern Kentucky. I would also recommend several steps in addition to the ideas put forth by McComb et al. (1991). First, establish a permanent system of monitoring that would include surveys of birds in all seral stages at low, mid-, and high elevations in the Cumberland Mountains and that would track species patterns over time as habitats mature or change due to land use practices and natural disturbance. Second, initiate research into nesting success of birds in disturbed and undisturbed habitats, with particular attention paid to causes of nest failure. Third, develop a Geographic Information Systems data base of habitats so that the frequency, block size, and distribution pattern of existing habitats could be identified. This information would allow for objective decision-making when allocating permits to log or mine, such that preservation of sufficient mature forest habitat to protect bird communities at low, mid-, and high elevations in the Cumberland Mountains could be achieved.

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# NOTES

**Seasonal Prevalence of the Digenetic Trematode *Proterometra edneyi* (Azygiidae) in the Snail *Elimia ebenum* (Pleuroceridae) at Anglin Falls, Kentucky.**—*Proterometra edneyi* is a digenetic trematode whose larval and adult stages were first described from the snail *Elimia semicarinata* and experimental and natural infections of several species of darters (*Etheostoma* spp.), respectively (1). To our knowledge, natural snail infections have been reported from streams in only seven Kentucky counties (1, 2, 3). The prevalence of *P. edneyi* infections in snail populations has been low at these sites, ranging from 0.45% at North Elkhorn Creek in Scott County (2) to 12.6% at South Elkhorn Creek in Fayette County (1). Little information is available concerning the seasonal prevalence of this worm. Uglem and Aliff (1) observed mature cercariae between March and October from their monthly (July 1980 to October 1981) collections of *Elimia semicarinata*, but no specific data regarding monthly prevalence were reported. Monthly prevalence can provide critical information regarding annual

loss/recruitment of trematodes within snail populations and optimal times for transmission to the next host in the worm's life cycle.

Our preliminary survey revealed the presence of *P. edneyi* rediae and cercariae in the snail *Elimia ebenum* and the adult worm in the striped darter *Etheostoma virgatum* at Anglin Falls, Rockcastle County, Kentucky. Our study was initiated to assess the seasonal prevalence of *P. edneyi* in the snail population at this site.

Anglin Falls is part of the Cumberland River drainage. The falls area, currently maintained by Berea College, is dedicated as a Kentucky State Nature Preserve encompassing 123 acres containing a number of intermittent streams. Beginning in June 1998, we collected 146–150 *Elimia ebenum* during the 4th week of each month through May 1999. Snails were placed individually into 50-ml beakers containing ca. 35–40 ml of filtered stream water. The beakers were then incubated in an environmental chamber at 20°C and a 12 hr light:12 hr dark cycle for 24 hr. Beakers were observed twice during this period

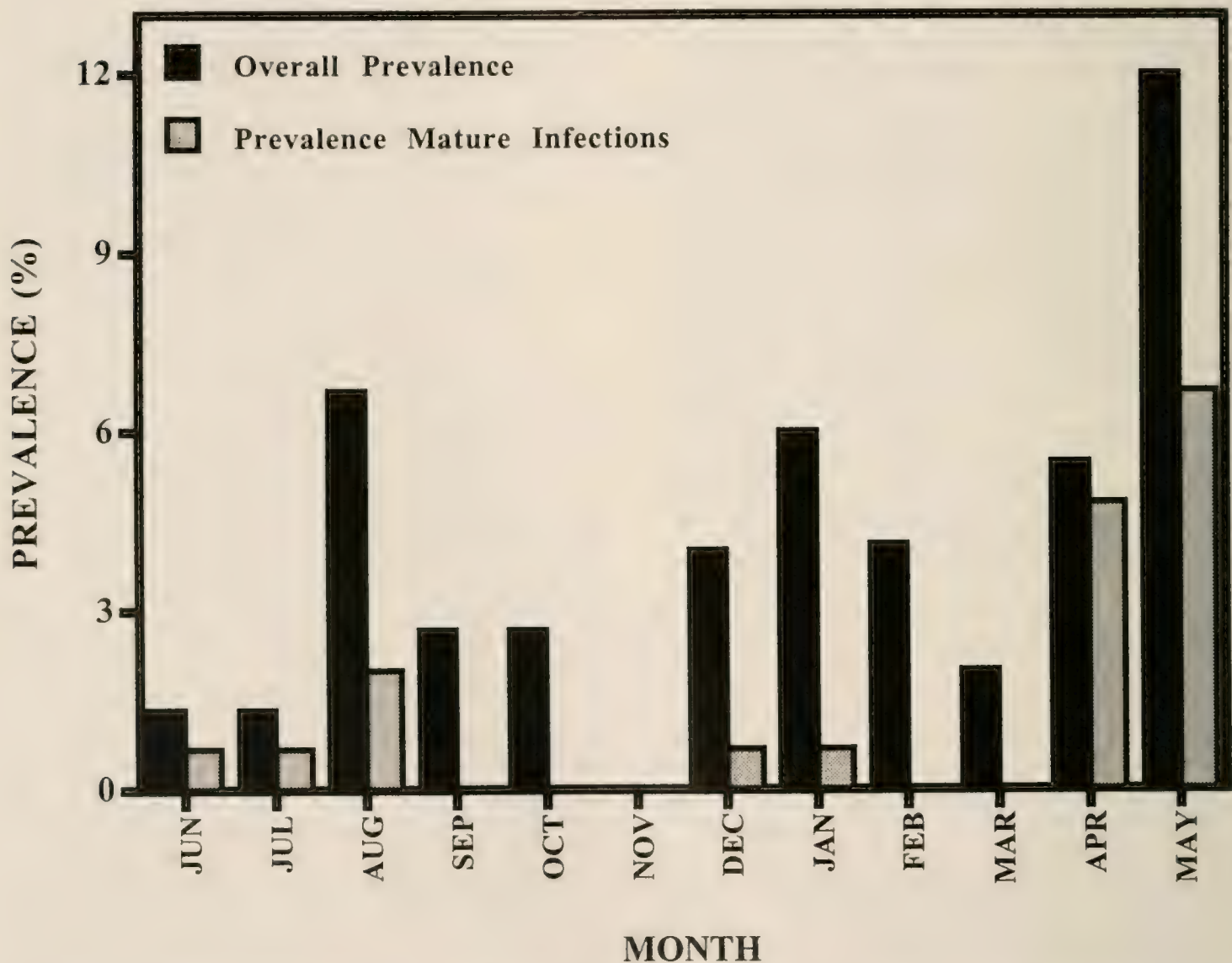


Figure 1. Monthly prevalence of *Proterometra edneyi* in the snail *Elimia ebenum* (solid box) and snails releasing mature cercariae of this species (stippled box) during June–May 1998–1999 at Anglin Falls, Kentucky.



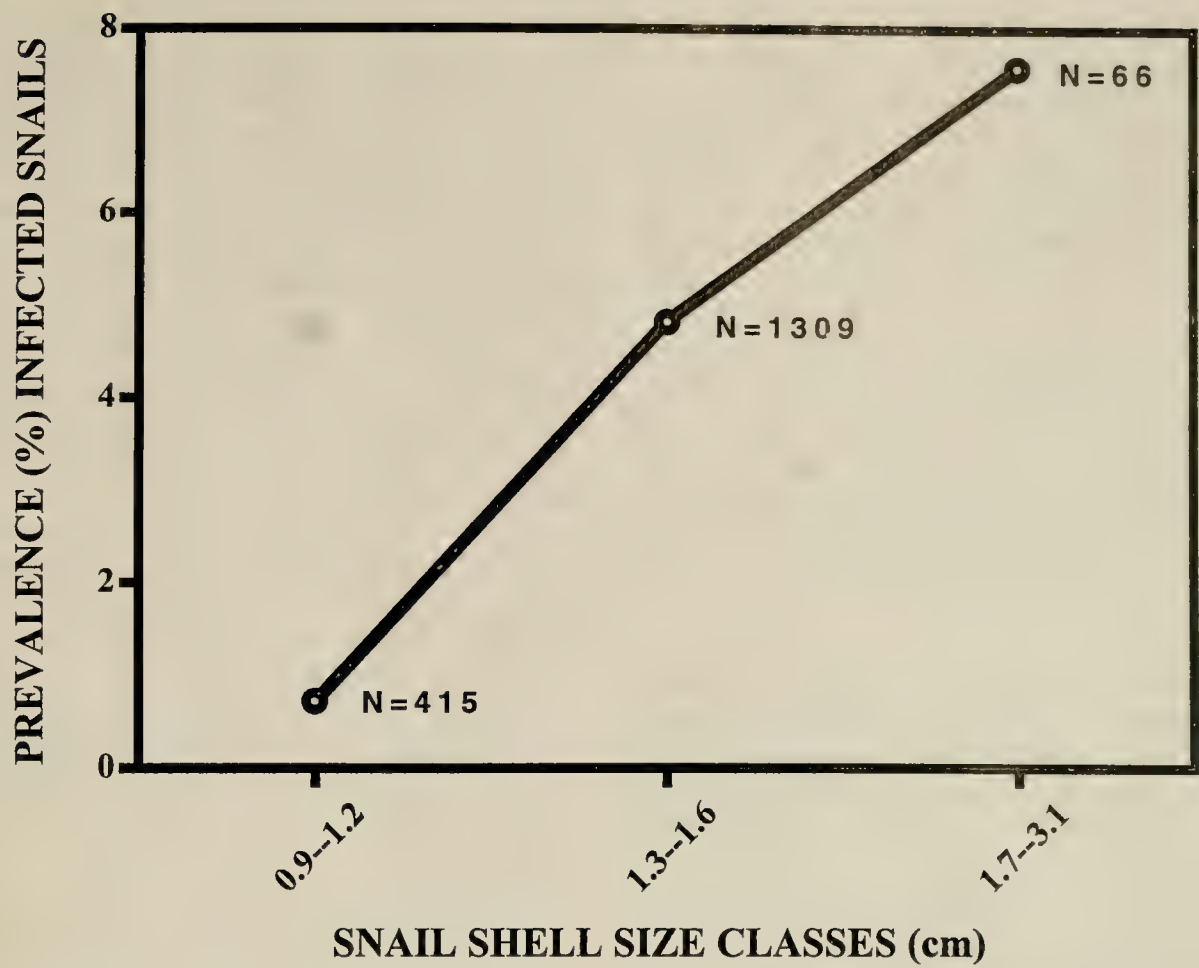


Figure 2. Prevalence of *Proterometra edneyi* in three size classes of the snail *Elimia ebenum* collected from June to May 1998–1999 at Anglin Falls, Kentucky. N = number of snails/size class.

with a dissecting microscope to determine which snails were releasing cercariae and thus possessed mature cercarial infections. Snail shell length was then recorded, and all snails were crushed to determine the presence/absence of rediae. Representative specimens were deposited in the U.S. National Parasite Collection (USNPC) with the following accession numbers: immature cercariae, 088844.0; cercariae, 088845.0; and rediae, 088846.0.

The 12-month prevalence of *P. edneyi* in *Elimia ebenum* at Anglin Falls was 4.02% (72/1790) in snails measuring between 0.9 and 3.1 cm. With the exception of November, prevalence revealed a continuous low-level infection in the snail population from June 1998 through May 1999 (prevalence range = 1.33–12.00%; Figure 1). There was a marked increase in the number of mature cercarial infections in April and May along with an overall increase in May prevalence (Figure 1). An increase in the prevalence of infection was also observed with increases in snail size (Figure 2).

The low prevalence of infection observed for *P. edneyi* in *Elimia ebenum* at Anglin Falls corroborates the observations made for this parasite in *Elimia semicarinata* (1, 2). Our observation of higher worm prevalence with increased snail size (Figure 2) also supports previous observations of *P. edneyi* infections in *Elimia semicarinata* (1). In samples of snails between 1.0 and 2.0 cm in length,

Uglen and Aliff (1) found the greatest prevalence in snails between 1.6 and 1.8 cm.

Based on the prevalence of mature cercarial infections, the primary period of *P. edneyi* transmission to *Etheostoma virgatum* at Anglin Falls must occur in spring. Riley and Uglen (4) speculated that seasonal emergence peaks of cercaria, are, in part, associated with the arrival of migrant hosts in their study of strains of the closely related species *Proterometra macrostoma*. According to Kuehne and Barbour (5), *Etheostoma virgatum* (which is common in second-, third-, and fourth order streams), sometimes enters the lower reaches of tiny woodland tributaries (like those found at Anglin Falls) in April to spawn. Such behavior would place this definitive host in close proximity with the mature/infective cercarial stage, which is most prevalent during this time (Figure 1). Further investigations into seasonal movements of this darter at Anglin Falls will be required to verify this association.

This study was supported by grants from the Andrew Mellon Foundation (Appalachian College Association) and the Undergraduate Research Creative Projects Program (URCPP) at Berea College. We acknowledge Dr. Guenter Schuster for reviewing the manuscript.

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**Chromosome Number of the Sandstone Rockhouse Endemic *Thalictrum mirabile* (Ranunculaceae), and Clarification of its Endemism.**—Sandstone rockhouses are semicircular recesses extending far back under cliff overhangs that are large enough to provide shelter for humans. Four ferns and seven flowering plants appear to be endemic, or nearly so, to sandstone rockhouses in the eastern United States (1, 2). The endemics have been classified following a cytologically based scheme: paleoendemic, neoschizoendemic, holoschizoendemic, patroendemic, or apoendemic (1, 3). A diploid or polyploid species with no apparent closely related extant diploid ancestor is a paleoendemic. Schizoendemics have the same chromosome number as their closely related parental taxa but are of various ages: geographically restricted, youthful species (neoschizoendemic) and widespread, “mature” or ancient species (holoschizoendemic). A restricted diploid species ancestral to a widespread polyploid is a patroendemic, whereas a restricted polyploid derived from a widespread diploid is an apoendemic.

*Thalictrum mirabile* Small (Ranunculaceae) was the only endemic flowering plant of the rockhouses that lacked a chromosome count, and thus it was classified tentatively as a neoschizoendemic (1). The purpose of my study was to (1) determine the chromosome number of *T. mirabile*, and (2) evaluate the species’ classification as a neoschizoendemic.

*Thalictrum mirabile* grows mostly around plunge basins and groundwater seeps/springs and at the heads of streams on the floor of rockhouses, and it is present on wet cliffs with slight overhangs (1, 4). The species was reported from Kentucky, Tennessee, North Carolina, Georgia, and Alabama by Park and Festerling (4). On the other hand, it is not listed for Tennessee by Wofford and Chester (5), North Carolina by Radford et al. (6), or Georgia by Jones and Coile (7). *Thalictrum mirabile* is very similar to its putative parental taxon, *T. clavatum* DC. The species are distinguished primarily by achene morphology (1, 4, 8). *Thalictrum clavatum* occurs in rich woods, on

cliffs and seepage slopes, and along streams from Virginia to Kentucky south to South Carolina and Georgia (4, 9).

Jensen (10) reported that *T. clavatum* from western North Carolina had a meiotic chromosome number of  $n = 7$ . The base chromosome number ( $x$ ) in *Thalictrum* is seven (8). Although Keener (9) included Jensen’s (10) chromosome count of *T. clavatum* in his treatment of *Thalictrum*, other recent taxonomic manuals (4, 8) have not. Moreover, the chromosome number of *T. clavatum* was omitted from Darlington and Wylie (11) and from Bolkhovskikh et al. (12), even though that of other species of *Thalictrum* in Jensen (10) was included in both sources.

I used young flower buds to determine the meiotic chromosome number of *T. mirabile* (cf. 13). Flower buds were collected from several genets in a population of *T. mirabile* in a rockhouse in Powell County, Kentucky, on 7 May 1999. A voucher specimen is deposited at OS (Walck 568). Plant material was placed in a 3:1 solution of absolute ethanol:glacial acetic acid for 2 days, and then transferred to 70% ethanol for 1 day. Anthers were removed from buds, placed in acetocarmine, macerated on a microscope slide, and then squashed with a cover slip. Slides were observed with a compound microscope, and chromosomes counted.

The chromosome number for *T. mirabile* was determined to be  $n = 7$ . This count is identical to that reported for *T. clavatum* (10). Thus, it is most appropriate to keep *T. mirabile* as a neoschizoendemic.

I thank Daniel J. Crawford for his guidance in this study.

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"Nauka," Leningrad. (13) Löve, Á., and D. Löve. 1975. Plant chromosomes. J. Cramer, Vaduz.—**Jeffrey L. Walck**, Department of Evolution, Ecology, and Organismal Biology, The Ohio State University, 1735 Neil Avenue, Columbus, OH 43210-1293; Present address: Department of Biology, P.O. Box 60, Middle Tennessee State University, Murfreesboro, TN 37132.



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